











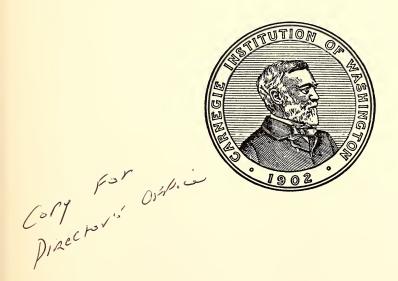




CARNEGIE INSTITUTION OF WASHINGTON

YEAR BOOK No. 51 July 1, 1951—June 30, 1952

With Administrative Reports through December 12, 1952



CARNEGIE INSTITUTION OF WASHINGTON WASHINGTON, D. C.
1952

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PRESIDENT AND TRUSTEES

PRESIDENT

Vannevar Bush

BOARD OF TRUSTEES

Elihu Root, Jr., Chairman LINDSAY BRADFORD, Vice-Chairman *Lewis H. Weed, Secretary

	*Lewis H. Weed, Secretary	
James F. Bell	Ernest O. Lawrence	HENNING W. PRENTIS, JR.
ROBERT WOODS BLISS	Alfred L. Loomis	David Rockefeller
LINDSAY BRADFORD	Robert A. Lovett	Elihu Root, Jr.
Omar N. Bradley	Keith S. McHugh	HENRY R. SHEPLEY
‡Homer L. Ferguson	Roswell Miller	CHARLES P. TAFT
W. CAMERON FORBES	HENRY S. MORGAN	JUAN T. TRIPPE
Crawford H. Greenewalt	Seeley G. Mudd	†JAMES W. WADSWORTH
CARYL P. HASKINS	WILLIAM I. MYERS	*Lewis H. Weed
BARKLIE HENRY		
	Executive Committee	
	Elihu Root, Jr., Chairman	
ROBERT WOODS BLISS	CARYL P. HASKINS	HENRY R. SHEPLEY
Vannevar Bush	Barklie Henry	*Lewis H. Weed
	HENNING W. PRENTIS, JR.	
	Finance Committee	
	LINDSAY BRADFORD, Chairman	
Alfred L. Loomis	Henry S. Morgan	Elihu Root, Jr.
THE RED L. LOOMIS	Henning W. Prentis, Jr.	Ellie Root, JR.
	Auditing Committee	
	_	
D II-	Roswell Miller, Chairman	VC M II
Barklie Henry	Maninatina Cammina	Кеітн S. МсНисн
	Nominating Committee	
D *** D	HENRY S. Morgan, Chairman	B
ROBERT WOODS BLISS	Robert A. Lovett	Elihu Root, Jr.
	Committee on Astronomy	
	Seeley G. Mudd, Chairman	
CRAWFORD H. GREENEWALT	*	Elihu Root, Jr.
		ELIHU KOOT, JR.
	Committee on Terrestrial Sciences	
	Ernest O. Lawrence, Chairman	
Barklie Henry		David Rockefeller
	Committee on Biological Sciences	
	*Lewis H. Weed, Chairman	
CARYL P. HASKINS	Alfred L. Loomis	CHARLES P. TAFT
	WILLIAM I. MYERS	
	Committee on Archaeology	
Large E. Pros	HENRY R. SHEPLEY, Chairman ROBERT WOODS BLISS	I T Trenns
James F. Bell	KOBERT WOODS BLISS	Juan T. Trippe

^{*} Deceased December 21, 1952. ‡ Resigned September 8, 1952. † Deceased June 21, 1952.

FORMER PRESIDENTS AND TRUSTEES

PRESIDENTS

Daniel Coit Gilman, 1902–1904 Robert Simpson Woodward, 1904–1920 John Campbell Merriam, *President* 1921–1938; *President Emeritus* 1939–1945

TRUSTEES

TREGITEE			
Alexander Agassiz	1904-05	Seth Low	1902–16
George J. Baldwin	1925–27	Wayne MacVeagh	1902-07
Thomas Barbour	1934–46	Andrew W. Mellon	1924-37
John S. Billings	1902-13	Darius O. Mills	1902-09
Robert S. Brookings	1910–29	S. Weir Mitchell	1902-14
John L. Cadwalader	1903-14	Andrew J. Montague	1907-35
WILLIAM W. CAMPBELL	1929–38	WILLIAM W. Morrow	1902–29
John J. Carty	1916–32	WILLIAM CHURCH OSBORN	1927-34
Whitefoord R. Cole	1925-34	James Parmelee	1917-31
Frederic A. Delano	1927-49	Wm. Barclay Parsons	1907-32
Cleveland H. Dodge	1903-23	Stewart Paton	1916–42
William E. Dodge	1902-03	George W. Pepper	1914-19
CHARLES P. FENNER	1914-24	John J. Pershing	1930-43
Homer L. Ferguson	1927-52	Henry S. Pritchett	1906–36
SIMON FLEXNER	1910-14	Gordon S. Rentschler	1946–48
JAMES FORRESTAL	1948–49	Ецни Коот	1902-37
WILLIAM N. FREW	1902-15	Julius Rosenwald	1929-31
Lyman J. Gage	1902-12	Martin A. Ryerson	1908–28
Walter S. Gifford	1931–50	Theobald Smith	1914-34
Cass Gilbert	1924-34	John C. Spooner	1902-07
Frederick H. Gillett	1924-35	William Benson Storey	1924-39
Daniel C. Gilman	1902–08	RICHARD P. STRONG	1934–48
John Hay	1902-05	William H. Taft	1906–15
Myron T. Herrick	1915-29	WILLIAM S. THAYER	1929–32
ABRAM S. HEWITT	1902-03	James W. Wadsworth	1932–52
HENRY L. HIGGINSON	1902-19	CHARLES D. WALCOTT	1902-27
Етнан А. Нітснсоск	1902-09	Frederic C. Walcott	1931–48°
Henry Hitchcock	1902-02	HENRY P. WALCOTT	1910–24
Herbert Hoover	1920-49	Lewis H. Weed	1935–52
William Wirt Howe	1903-09	WILLIAM H. WELCH	1906–34
Charles L. Hutchinson	1902-04	Andrew D. White	1902-03
Walter A. Jessup	1938–44	Edward D. White	1902-03
Frank B. Jewett	1933-49	HENRY WHITE	1913-27
SAMUEL P. LANGLEY	1904–06	George W. Wickersham	1909–36
Charles A. Lindbergh	1934-39	Robert S. Woodward	1905–24
WILLIAM LINDSAY	1902-09	Carroll D. Wright	1902–08
HENRY CABOT LODGE	1914-24		

Besides the names enumerated above, the following were ex-officio members of the Board of Trustees under the original charter, from the date of organization until April 28, 1904: the President of the United States, the President of the Senate, the Speaker of the House of Representatives, the Secretary of the Smithsonian Institution, the President of the National Academy of Sciences.

STAFF OF INVESTIGATORS FOR THE YEAR 1952

ASTRONOMY

MOUNT WILSON AND PALOMAR OBSERVATORIES

813 Santa Barbara Street, Pasadena 4, California

Mount Wilson Observatory organized in 1904; George E. Hale, Director 1904–1923, Honorary Director 1923–1936; Walter S. Adams, Director 1924–1945. Unified operation with the Palomar Observatory of the California Institute of Technology began in 1948.

IRA S. BOWEN, Director
WALTER BAADE
HORACE W. BABCOCK
WILLIAM A. BAUM
ARMIN J. DEUTSCH
JESSE L. GREENSTEIN
EDWIN P. HUBBLE
MILTON L. HUMASON

PAUL W. MERRILL

RUDOLPH L. MINKOWSKI
GUIDO MÜNCH
SETH B. NICHOLSON
EDISON PETTIT
ROBERT S. RICHARDSON
ALLAN R. SANDAGE
ALBERT G. WILSON
OLIN C. WILSON

FRITZ ZWICKY

TERRESTRIAL SCIENCES

GEOPHYSICAL LABORATORY

2801 Upton Street, N.W., Washington 8, D. C.

Organized in 1906, opened in 1907; Arthur L. Day, Director 1909–1936; Leason H. Adams, Acting Director 1936–1937, Director 1938—August 1, 1952.

*Leason H. Adams, Director
George W. Morey, Acting Director,
August 1, 1952
*Norman L. Bowen
‡John S. Burlew
Felix Chayes
Gordon L. Davis
Joseph L. England

JOSEPH W. GREIG FRANK C. KRACEK J. FRANK SCHAIRER O. FRANK TUTTLE HATTEN S. YODER

Visiting Investigator
HANS P. EUGSTER

DEPARTMENT OF TERRESTRIAL MAGNETISM

5241 Broad Branch Road, N.W., Washington 15, D. C.

Organized in 1904; Louis A. Bauer, Director 1904–1929; John A. Fleming, Acting Director 1929–1934, Director 1935–1946.

MERLE A. TUVE, Director
PHILIP H. ABELSON
L. THOMAS ALDRICH
ELLIS T. BOLTON
ROY J. BRITTEN
DEAN B. COWIE
SCOTT E. FORBUSH
JOHN W. GRAHAM
NORMAN P. HEYDENBURG
§ELLIS A. JOHNSON
RICHARD B. ROBERTS
HOWARD E. TATEL

GEORGE R. TILTON

OSCAR W. TORRESON

ERNEST H. VESTINE

HARRY W. WELLS

Visiting Investigators
William R. Duryee
||John W. Findlay
Pembroke J. Hart
||Soren Lövtrup

KENNETH R. McQuillan

||A. T. Price ||Gunnar Randers | Irena Z. Roberts | Georges M. Temmer

*Retired in 1952. ‡Resigned in 1952. §On leave of absence.

Term of appointment completed in 1952.

CARNEGIE INSTITUTION OF WASHINGTON

BIOLOGICAL SCIENCES

DEPARTMENT OF PLANT BIOLOGY

Stanford, California

Desert Laboratory, opened in 1903, became headquarters of Department of Botanical Research in 1905; name changed to Laboratory for Plant Physiology in 1923; Daniel T. MacDougal, Director 1906–1927. Reorganized in 1928 as Division of Plant Biology, including Ecology; Herman A. Spochr, Chairman 1927–1930 and 1931–1947, Chairman Emeritus 1947–1950. Name changed to Department of Plant Biology in 1951.

C. STACY FRENCH, Director

JENS C. CLAUSEN
PAUL GRUN

WILLIAM M. HIESEY HAROLD W. MILNER

Malcolm A. Nobs

JAMES H. C. SMITH

VIOLET K. YOUNG

Guest Investigators

FRIEDRICH EHRENDORFER

AXEL NYGREN

Visiting Investigator

JACK E. MYERS

DEPARTMENT OF EMBRYOLOGY

Wolfe and Madison Streets, Baltimore 5, Maryland

Organized in 1914; Franklin P. Mall, Director 1914-1917; George L. Streeter, Director 1918-1940

*George W. Corner, Director

SAMUEL R. M. REYNOLDS, Acting Director

Bent G. Böving

ROBERT K. BURNS ARPAD CSAPO Consultant

GEORGE W. BARTELMEZ

Research Associate

ELIZABETH M. RAMSEY

DEPARTMENT OF GENETICS

Cold Spring Harbor, Long Island, New York

Station for Experimental Evolution opened in 1904; name changed to Department of Experimental Evolution in 1906; combined with Eugenics Record Office in 1921 to form Department of Genetics. Charles B. Davenport, Director 1904–1934; Albert F. Blakeslee, Director 1935–1941.

MILISLAV DEMEREC, Director

ALFRED D. HERSHEY BERWIND P. KAUFMANN

‡Edwin C. MacDowell

BARBARA McCLINTOCK

MARGARET R. McDonald

N. VISCONTI

EVELYN M. WITKIN

Visiting Investigator

M. J. D. WHITE

ARCHAEOLOGY

DEPARTMENT OF ARCHAEOLOGY

10 Frisbie Place, Cambridge 38, Massachusetts

Department of Historical Research organized in 1903; Andrew C. McLaughlin, Director 1903–1905; J. Franklin Jameson, Director 1905–1928. In 1930 this Department was incorporated as a section of United States history in a new Division of Historical Research; Alfred V. Kidder, Chairman 1930–1950. Name changed to Department of Archaeology in 1951.

HARRY E. D. POLLOCK, Director

MARGARET W. HARRISON, Editor

EARL H. MORRIS

TATIANA PROSKOURIAKOFF

KARL RUPPERT

Anna O. Shepard

EDWIN M. SHOOK

A. LEDYARD SMITH

ROBERT E. SMITH

Gustav Strömsvik

J. ERIC S. THOMPSON

* On leave of absence. ‡ Retired in 1952.

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STAFF OF INVESTIGATORS FOR THE YEAR 1952

RESEARCH ASSOCIATES

RESEARCH ASSOCIATES ENGAGED IN POST-RETIREMENT STUDIES

CHESTER H. HEUSER, Embryology

RALPH L. Roys, History

RESEARCH ASSOCIATES CONNECTED WITH OTHER INSTITUTIONS

RALPH W. CHANEY (University of California), Paleobotany Louis B. Flexner (University of Pennsylvania), Embryology Arthur T. Hertig (Boston Lying-in Hospital), Embryology E. A. Lowe (The Institute for Advanced Study), Paleography

OFFICE OF ADMINISTRATION

Vannevar Bush, President
Paul A. Scherer, Executive Officer
Samuel Callaway, Assistant to the President
Ailene J. Bauer, Director of Publications
Dorothy R. Swift, Editor
John A. Fleming, Adviser in International Scientific Relations
Earle B. Biesecker, Bursar
J. Stanley Lingebach, Assistant Bursar
James F. Sullivan, Assistant to the Bursar
Richard F. F. Nichols, Executive Assistant to the Finance Committee

ORGANIZATION, PLAN, AND SCOPE

The Carnegie Institution of Washington was founded by Andrew Carnegie, January 28, 1902, when he gave to a board of trustees an endowment of registered bonds of the par value of ten million dollars. To this fund an addition of two million dollars was made by Mr. Carnegie on December 10, 1907, and a further addition of ten million dollars was made by him on January 19, 1911. Furthermore, the income of a reserve fund of about three million dollars, accumulated in accordance with the founder's specifications in 1911, is now available for general use, and in recent years a total of ten million dollars has been paid by the Carnegie Corporation of New York as increase to the Endowment Fund of the Institution. The Institution was originally organized under the laws of the District of Columbia and incorporated as the *Carnegie Institution*, articles of incorporation having been executed on January 4, 1902. The Institution was reincorporated, however, by an act of the Congress of the United States, approved April 28, 1904, under the title of *Carnegie Institution of Washington*. (See existing Articles of Incorporation on following pages.)

Organization under the new Articles of Incorporation was effected May 18, 1904, and the Institution was placed under the control of a board of twenty-four trustees, all of whom had been members of the original corporation. The trustees meet annually in December to consider the affairs of the Institution in general, the progress of work already undertaken, and the initiation of new projects, and to make the necessary appropriations for the ensuing year. During the intervals between the meetings of the trustees the affairs of the Institution are conducted by an Executive Committee chosen by and from the Board of Trustees and acting through the President of the Institution as chief executive officer.

The Articles of Incorporation of the Institution declare in general "that the objects of the corporation shall be to encourage, in the broadest and most liberal manner, investigation, research, and discovery, and the application of knowledge to the improvement of mankind."

The Institution is essentially an operating organization. It attempts to advance fundamental research in fields not normally covered by the activities of other agencies, and to concentrate its attention upon specific problems, with the idea of shifting attack from time to time to meet more pressing needs of research as they develop with increase of knowledge. Some of these problems require the collaboration of several investigators, special equipment, and continuous effort. Many close relations exist among activities of the Institution, and a type of organization representing investigations in astronomy, in terrestrial sciences, in biological sciences, and in archaeology has been effected. Conference groups on various subjects have played a part in bringing new vision and new methods to bear upon many problems. Constant efforts are made to facilitate interpretation and application of results of research activities of the Institution.

ARTICLES OF INCORPORATION

Public No. 260. An Act to incorporate the Carnegie Institution of Washington.

Be it enacted by the Senate and House of Representatives of the United States of America in Congress assembled, That the persons following, being persons who are now trustees of the Carnegie Institution, namely, Alexander Agassiz, John S. Billings, John L. Cadwalader, Cleveland H. Dodge, William N. Frew, Lyman J. Gage, Daniel C. Gilman, John Hay, Henry L. Higginson, William Wirt Howe, Charles L. Hutchinson, Samuel P. Langley, William Lindsay, Seth Low, Wayne MacVeagh, Darius O. Mills, S. Weir Mitchell, William W. Morrow, Ethan A. Hitchcock, Elihu Root, John C. Spooner, Andrew D. White, Charles D. Walcott, Carroll D. Wright, their associates and successors, duly chosen, are hereby incorporated and declared to be a body corporate by the name of the Carnegie Institution of Washington and by that name shall be known and have perpetual succession, with the powers, limitations, and restrictions herein contained.

- SEC. 2. That the objects of the corporation shall be to encourage, in the broadest and most liberal manner, investigation, research, and discovery, and the application of knowledge to the improvement of mankind; and in particular—
- (a) To conduct, endow, and assist investigation in any department of science, literature, or art, and to this end to cooperate with governments, universities, colleges, technical schools, learned societies, and individuals.
 - (b) To appoint committees of experts to direct special lines of research.
 - (c) To publish and distribute documents.
 - (d) To conduct lectures, hold meetings, and acquire and maintain a library.
- (e) To purchase such property, real or personal, and construct such building or buildings as may be necessary to carry on the work of the corporation.
- (f) In general, to do and perform all things necessary to promote the objects of the institution, with full power, however, to the trustees hereinafter appointed and their successors from time to time to modify the conditions and regulations under which the work shall be carried on, so as to secure the application of the funds in the manner best adapted to the conditions of the time, provided that the objects of the corporation shall at all times be among the foregoing or kindred thereto.
- Sec. 3. That the direction and management of the affairs of the corporation and the control and disposal of its property and funds shall be vested in a board of trustees, twenty-two in number, to be composed of the following individuals: Alexander Agassiz, John S. Billings, John L. Cadwalader, Cleveland H. Dodge, William N. Frew, Lyman J. Gage, Daniel C. Gilman, John Hay, Henry L. Higginson, William Wirt Howe, Charles L. Hutchinson, Samuel P. Langley, William Lindsay, Seth Low, Wayne MacVeagh, Darius O. Mills, S. Weir Mitchell, William W. Morrow, Ethan A. Hitchcock, Elihu Root, John C. Spooner, Andrew D. White, Charles D. Walcott, Carroll D. Wright, who shall constitute the first board of trustees. The board of trustees shall have power from time to time to increase its membership to not more than twenty-seven members. Vacancies occasioned by death, resignation, or otherwise shall be filled by the remaining trustees in such manner as the by-laws

ARTICLES OF INCORPORATION

shall prescribe; and the persons so elected shall thereupon become trustees and also members of the said corporation. The principal place of business of the said corporation shall be the city of Washington, in the District of Columbia.

Sec. 4. That such board of trustees shall be entitled to take, hold, and administer the securities, funds, and property so transferred by said Andrew Carnegie to the trustees of the Carnegie Institution and such other funds or property as may at any time be given, devised, or bequeathed to them, or to such corporation, for the purposes of the trust; and with full power from time to time to adopt a common seal, to appoint such officers, members of the board of trustees or otherwise, and such employees as may be deemed necessary in carrying on the business of the corporation, at such salaries or with such remuneration as they may deem proper; and with full power to adopt by-laws from time to time and such rules or regulations as may be necessary to secure the safe and convenient transaction of the business of the corporation; and with full power and discretion to deal with and expend the income of the corporation in such manner as in their judgment will best promote the objects herein set forth and in general to have and use all powers and authority necessary to promote such objects and carry out the purposes of the donor. The said trustees shall have further power from time to time to hold as investments the securities hereinabove referred to so transferred by Andrew Carnegie, and any property which has been or may be transferred to them or such corporation by Andrew Carnegie or by any other person, persons, or corporation, and to invest any sums or amounts from time to time in such securities and in such form and manner as are permitted to trustees or to charitable or literary corporations for investment, according to the laws of the States of New York, Pennsylvania, or Massachusetts, or in such securities as are authorized for investment by the said deed of trust so executed by Andrew Carnegie, or by any deed of gift or last will and testament to be hereafter made or executed.

SEC. 5. That the said corporation may take and hold any additional donations, grants, devises, or bequests which may be made in further support of the purposes of the said corporation, and may include in the expenses thereof the personal expenses which the trustees may incur in attending meetings or otherwise in carrying out the business of the trust, but the services of the trustees as such shall be gratuitous.

SEC. 6. That as soon as may be possible after the passage of this Act a meeting of the trustees hereinbefore named shall be called by Daniel C. Gilman, John S. Billings, Charles D. Walcott, S. Weir Mitchell, John Hay, Elihu Root, and Carroll D. Wright, or any four of them, at the city of Washington, in the District of Columbia, by notice served in person or by mail addressed to each trustee at his place of residence; and the said trustees, or a majority thereof, being assembled, shall organize and proceed to adopt by-laws, to elect officers and appoint committees, and generally to organize the said corporation; and said trustees herein named, on behalf of the corporation hereby incorporated, shall thereupon receive, take over, and enter into possession, custody, and management of all property, real or personal, of the corporation heretofore known as the Carnegie Institution, incorporated, as hereinbefore set forth under "An Act to establish a Code of Law for the District of Columbia, January fourth, nineteen hundred and two," and to all its rights, contracts, claims, and property of any kind or nature; and the several officers of such corporation, or

CARNEGIE INSTITUTION OF WASHINGTON

any other person having charge of any of the securities, funds, real or personal, books, or property thereof, shall, on demand, deliver the same to the said trustees appointed by this Act or to the persons appointed by them to receive the same; and the trustees of the existing corporation and the trustees herein named shall and may take such other steps as shall be necessary to carry out the purposes of this Act.

Sec. 7. That the rights of the creditors of the said existing corporation known as the Carnegie Institution shall not in any manner be impaired by the passage of this Act, or the transfer of the property hereinbefore mentioned, nor shall any liability or obligation for the payment of any sums due or to become due, or any claim or demand, in any manner or for any cause existing against the said existing corporation, be released or impaired; but such corporation hereby incorporated is declared to succeed to the obligations and liabilities and to be held liable to pay and discharge all of the debts, liabilities, and contracts of the said corporation so existing to the same effect as if such new corporation had itself incurred the obligation or liability to pay such debt or damages, and no such action or proceeding before any court or tribunal shall be deemed to have abated or been discontinued by reason of the passage of this Act.

SEC. 8. That Congress may from time to time alter, repeal, or modify this Act of incorporation, but no contract or individual right made or acquired shall thereby be divested or impaired.

Sec. 9. That this Act shall take effect immediately.

Approved, April 28, 1904

BY-LAWS OF THE INSTITUTION

Adopted December 13, 1904. Amended December 13, 1910, December 13, 1912, December 10, 1937, December 15, 1939, December 13, 1940, December 18, 1942, and December 12, 1947

ARTICLE I

THE TRUSTEES

- 1. The Board of Trustees shall consist of twenty-four members, with power to increase its membership to not more than twenty-seven members. The Trustees shall hold office continuously and not for a stated term.
- 2. In case any Trustee shall fail to attend three successive annual meetings of the Board he shall thereupon cease to be a Trustee.
 - 3. No Trustee shall receive any compensation for his services as such.
- 4. All vacancies in the Board of Trustees shall be filled by the Trustees by ballot at an annual meeting, but no person shall be declared elected unless he receives the votes of two-thirds of the Trustees present.

ARTICLE II

OFFICERS OF THE BOARD

- I. The officers of the Board shall be a Chairman of the Board, a Vice-Chairman, and a Secretary, who shall be elected by the Trustees, from the members of the Board, by ballot to serve for a term of three years. All vacancies shall be filled by the Board for the unexpired term; provided, however, that the Executive Committee shall have power to fill a vacancy in the office of Secretary to serve until the next meeting of the Board of Trustees.
- 2. The Chairman shall preside at all meetings and shall have the usual powers of a presiding officer.
- 3. The Vice-Chairman, in the absence or disability of the Chairman, shall perform the duties of the Chairman.
- 4. The Secretary shall issue notices of meetings of the Board, record its transactions, and conduct that part of the correspondence relating to the Board and to his duties.

ARTICLE III

EXECUTIVE ADMINISTRATION

The President

I. There shall be a President who shall be elected by ballot by, and hold office during the pleasure of, the Board, who shall be the chief executive officer of the Institution. The President, subject to the control of the Board and the Executive Committee, shall have general charge of all matters of administration and supervision of all arrangements for research and other work undertaken by the Institution or with its funds. He shall prepare and submit to the Board of Trustees and to the Executive

CARNEGIE INSTITUTION OF WASHINGTON

Committee plans and suggestions for the work of the Institution, shall conduct its general correspondence and the correspondence with applicants for grants and with the special advisers of the Committee, and shall present his recommendations in each case to the Executive Committee for decision. All proposals and requests for grants shall be referred to the President for consideration and report. He shall have power to remove, appoint, and, within the scope of funds made available by the Trustees, provide for compensation of subordinate employees and to fix the compensation of such employees within the limits of a maximum rate of compensation to be established from time to time by the Executive Committee. He shall be *ex officio* a member of the Executive Committee.

- 2. He shall be the legal custodian of the seal and of all property of the Institution whose custody is not otherwise provided for. He shall sign and execute on behalf of the corporation all contracts and instruments necessary in authorized administrative and research matters and affix the corporate seal thereto when necessary, and may delegate the performance of such acts and other administrative duties in his absence to the Executive Officer. He may execute all other contracts, deeds, and instruments on behalf of the corporation and affix the seal thereto when expressly authorized by the Board of Trustees or Executive Committee. He may, within the limits of his own authorization, delegate to the Executive Officer authority to act as custodian of and affix the corporate seal. He shall be responsible for the expenditure and disbursement of all funds of the Institution in accordance with the directions of the Board and of the Executive Committee, and shall keep accurate accounts of all receipts and disbursements. Following approval by the Executive Committee he shall transmit to the Board of Trustees before its annual meeting in December a written report of the operations and business of the Institution for the preceding fiscal year with his recommendations for work and appropriations for the succeeding calendar year.
 - 3. He shall attend all meetings of the Board of Trustees.
- 4. There shall be an officer designated Executive Officer who shall be appointed by and hold office at the pleasure of the President, subject to the approval of the Executive Committee. His duties shall be to assist and act for the President as the latter may duly authorize and direct.
- 5. The President shall retire from office at the end of the calendar year in which he becomes sixty-five years of age.

ARTICLE IV

MEETINGS

- I. The annual meeting of the Board of Trustees shall be held in the City of Washington, in the District of Columbia, on the first Friday following the second Thursday of December in each year unless the date and place of meeting are otherwise ordered by the Executive Committee.
- 2. Special meetings of the Board may be called by the Executive Committee by notice served personally upon, or mailed to the usual address of, each Trustee twenty days prior to the meeting.
- 3. Special meetings shall, moreover, be called in the same manner by the Chairman upon the written request of seven members of the Board.

ARTICLE V

COMMITTEES

- 1. There shall be the following standing Committees, viz. an Executive Committee, a Finance Committee, an Auditing Committee, and a Nominating Committee.
- 2. All vacancies occurring in the Executive Committee, the Finance Committee, the Auditing Committee, and the Nominating Committee shall be filled by the Trustees at the next regular meeting. In case of vacancy in the Finance Committee, the Auditing Committee, or the Nominating Committee, upon request of the remaining members of such committee, the Executive Committee may fill such vacancy by appointment until the next meeting of the Board of Trustees.
- 3. The terms of all officers and of all members of committees, as provided for herein, shall continue until their successors are elected or appointed.

Executive Committee

- 4. The Executive Committee shall consist of the Chairman and Secretary of the Board of Trustees and the President of the Institution *ex officio* and, in addition, five trustees to be elected by the Board by ballot for a term of three years, who shall be eligible for re-election. Any member elected to fill a vacancy shall serve for the remainder of his predecessor's term.
- 5. The Executive Committee shall, when the Board is not in session and has not given specific directions, have general control of the administration of the affairs of the corporation and general supervision of all arrangements for administration, research, and other matters undertaken or promoted by the Institution. It shall also submit to the Board of Trustees a printed or typewritten report of each of its meetings, and at the annual meeting shall submit to the Board a report for publication.
- 6. The Executive Committee shall have power to authorize the purchase, sale, exchange, or transfer of real estate.

Finance Committee

- 7. The Finance Committee shall consist of five members to be elected by the Board of Trustees by ballot for a term of three years.
- 8. The Finance Committee shall have custody of the securities of the corporation and general charge of its investments and invested funds, and shall care for and dispose of the same subject to the directions of the Board of Trustees. It shall have power to authorize the purchase, sale, exchange, or transfer of securities and to delegate this power. It shall consider and recommend to the Board from time to time such measures as in its opinion will promote the financial interests of the Institution, and shall make a report at each meeting of the Board.

Auditing Committee

9. The Auditing Committee shall consist of three members to be elected by the Board of Trustees by ballot for a term of three years.

10. Before each annual meeting of the Board of Trustees, the Auditing Committee shall cause the accounts of the Institution for the preceding fiscal year to be audited by public accountants. The accountants shall report to the Committee, and the Committee shall present said report at the ensuing annual meeting of the Board with such recommendations as the Committee may deem appropriate.

Nominating Committee

- Trustees ex officio and, in addition, three trustees to be elected by the Board by ballot for a term of three years, who shall not be eligible for re-election until after the lapse of one year. Any member elected to fill a vacancy shall serve for the remainder of his predecessor's term, provided that of the Nominating Committee first elected after adoption of this By-Law one member shall serve for one year, one member shall serve for two years, and one member shall serve for three years, the Committee to determine the respective terms by lot.
- 12. Sixty days prior to an annual meeting of the Board the Nominating Committee shall notify the Trustees by mail of the vacancies to be filled in membership of the Board. Each Trustee may submit nominations for such vacancies. Nominations so submitted shall be considered by the Nominating Committee, and ten days prior to the annual meeting the Nominating Committee shall submit to members of the Board by mail a list of the persons so nominated, with its recommendations for filling existing vacancies on the Board and its Standing Committees. No other nominations shall be received by the Board at the annual meeting except with the unanimous consent of the Trustees present.

ARTICLE VI

FINANCIAL ADMINISTRATION

- 1. No expenditure shall be authorized or made except in pursuance of a previous appropriation by the Board of Trustees, or as provided in Article V, paragraph 8, hereof.
- 2. The fiscal year of the Institution shall commence on the first day of July in each year.
- 3. The Executive Committee shall submit to the annual meeting of the Board a full statement of the finances and work of the Institution for the preceding fiscal year and a detailed estimate of the expenditures of the succeeding calendar year.
- 4. The Board of Trustees, at the annual meeting in each year, shall make general appropriations for the ensuing calendar year; but nothing contained herein shall prevent the Board of Trustees from making special appropriations at any meeting.
- 5. The Executive Committee shall have general charge and control of all appropriations made by the Board. Following the annual meeting each year, the Executive Committee may make allotment of funds for the period from January 1 to termination of the fiscal year on June 30. It may also make allotment of funds for the period from July 1 to December 31 in advance of July 1. The Committee shall, however, have full authority for allotment of available funds to meet necessary

BY-LAWS OF THE INSTITUTION

expenditures by other methods, if desirable, and for transfer of balances to meet special needs. It shall make provision for outstanding obligations and for revertment of unexpended balances at termination of the fiscal year.

- 6. The securities of the Institution and evidences of property, and funds invested and to be invested, shall be deposited in such safe depository or in the custody of such trust company and under such safeguards as the Finance Committee shall designate, subject to directions of the Board of Trustees. Income of the Institution available for expenditure shall be deposited in such banks or depositories as may from time to time be designated by the Executive Committee.
- 7. Any trust company entrusted with the custody of securities by the Finance Committee may, by resolution of the Board of Trustees, be made Fiscal Agent of the Institution, upon an agreed compensation, for the transaction of the business coming within the authority of the Finance Committee.

ARTICLE VII

AMENDMENT OF BY-LAWS

1. These by-laws may be amended at any annual or special meeting of the Board of Trustees by a two-thirds vote of the members present, provided written notice of the proposed amendment shall have been served personally upon, or mailed to the usual address of, each member of the Board twenty days prior to the meeting.



ABSTRACT OF MINUTES OF THE FIFTY-FOURTH MEETING OF THE BOARD OF TRUSTEES

The annual meeting of the Board of Trustees was held in Washington, D. C., in the Board Room of the Administration Building, on Friday, December 12, 1952. It was called to order at 10:38 A.M. by the Chairman, Mr. Root.

The following Trustees were in attendance: Robert Woods Bliss, Lindsay Bradford, W. Cameron Forbes, Caryl P. Haskins, Barklie Henry, Ernest O. Lawrence, Keith S. McHugh, Roswell Miller, Henry S. Morgan, William I. Myers, Henning W. Prentis, Jr., Elihu Root, Jr., Henry R. Shepley, Charles P. Taft, and Juan T. Trippe. The President of the Institution, Vannevar Bush, was present.

The minutes of the fifty-third meeting were approved.

Reports of the President, the Executive Committee, the Finance Committee, the Auditor, the Auditing Committee, and the Nominating Committee, and of the Directors of Departments and Research Associates of the Institution were presented and considered.

The sum of \$1,842,595 was appropriated for the calendar year 1953 for expenditure under the general charge and control of the Executive Committee.

The Chairman announced the death of James W. Wadsworth and the resignation of Homer L. Ferguson during the past year. Crawford H. Greenewalt, of Wilmington, Delaware, and David Rockefeller, of New York, N. Y., were elected members of the Board of Trustees to fill these vacancies. By unanimous consent and action, Omar N. Bradley and Robert A. Lovett were re-elected members of the Board.

Henning W. Prentis, Jr. was re-elected a member of the Executive Committee for a three-year term. Barklie Henry and Keith S. McHugh were elected members of the Auditing Committee for three-year terms, to fill the vacancies on this Committee due to the death of James W. Wadsworth and the resignation of Homer L. Ferguson. Robert A. Lovett was elected a member of the Nominating Committee for a three-year term, succeeding Lindsay Bradford.

The meeting adjourned at 12:15 P.M.



REPORT OF THE EXECUTIVE COMMITTEE

To the Trustees of the Carnegie Institution of Washington:

Gentlemen: In accordance with the provisions of the By-Laws, the Executive Committee submits this report to the annual meeting of the Board of Trustees.

During the fiscal year ended June 30, 1952, the Executive Committee held four meetings, printed reports of which have been mailed to each Trustee and constitute a part of this report.

The detailed record of the activities of the Institution is presented in the reports from the Departments, which are contained in the Year Book, a review of some of the highlights being given in the report of the President. The estimate of expenditures for the calendar year 1953 contained in the report of the President has been considered by the Executive Committee, and the Committee recommends to the Board the adoption of the proposed budget based thereon.

The Board of Trustees, at its meeting of December 14, 1951, appointed the firm of Haskins & Sells to audit the accounts of the Institution for the fiscal year ending June 30, 1952. The report of the Auditor, including a balance sheet showing assets and liabilities of the Institution on June 30, 1952, together with supporting statements and schedules, is submitted as a part of the report of the Executive Committee.

Three vacancies exist in the membership of the Board of Trustees, resulting from the death in June 1952 of James W. Wadsworth, the previously reported resignation of Walter S. Gifford, and the recent resignation of Homer L. Ferguson. The resignation of Mr. Ferguson, received in September 1952, has been accepted by the Executive Committee.

The term of Mr. Prentis as a member of the Executive Committee and the term of Mr. Bradford as a member of the Nominating Committee will end at the annual meeting. Two vacancies have occurred in the Auditing Committee by reason of the death of Mr. Wadsworth and the resignation of Mr. Ferguson. The Executive Committee at its meeting on October 16, 1952 appointed Barklie Henry and Keith S. McHugh to fill the vacancies in the Auditing Committee until the annual meeting of the Board of Trustees in December 1952.

ELIHU ROOT, JR., Chairman

October 16, 1952



HASKINS & SELLS CERTIFIED PUBLIC ACCOUNTANTS

500 Equitable Building Baltimore 2

ACCOUNTANTS' CERTIFICATE

To the Board of Trustees of Carnegie Institution of Washington:

We have examined the balance sheet of Carnegie Institution of Washington as of June 30, 1952 and the related statements of income and expenditures and current funds surplus and the summaries of changes in endowment and other special funds and changes in investment in real estate and equipment for the year then ended (Exhibits A to E, inclusive). Our examination was made in accordance with generally accepted auditing standards, and accordingly included such tests of the accounting records and such other auditing procedures as we considered necessary in the circumstances, except that we did not examine the records in support of expenditures made (approximately \$262,000) by five of the seven branch offices of the Institution, but we have reviewed internal audit reports of the Bursar's office covering examinations of all branch records during the year.

In our opinion, subject to the exception stated above with respect to the limitation of the scope of our examination, the accompanying balance sheet and statements of income and expenditures and current funds surplus and summaries of changes in endowment and other special funds and changes in investment in real estate and equipment (Exhibits A to E, inclusive) present fairly the financial position of the Institution at June 30, 1952 and the results of its operations for the year then ended, in conformity with generally accepted accounting principles applied on a basis consistent with that of the preceding year.

HASKINS & SELLS

August 29, 1952

Balance Sheet, June 30, 1952

Assets

Current Funds:			
Cash in banks and on hand		\$683,006.09	
Advances:		14.040.04	
Departmental Research Operations		16,362.91	
Other	• • • • • • • • • • • • • • • •	430.00	
U. S. Government		343.94	
Other		349.99	
Inventory of books		108,937.40	
Deferred charges		29,487.69	
Due from Endowment and Other Special Fund		218,647.00	
·			, ,
Endowment and Other Special Funds:			
Cash in banks		\$122,175.65	
Securities (valuation based on market quota-		Ψ122,175.05	
tions at June 30, 1952, \$52,546,226),			
per Schedule 1:			
Bonds	\$22,977,299.06		
Preferred stocks	4,238,591.05		
Common stocks	16,174,393.61	43,390,283.72	43,512,459.37
Plant Funds:			
Investment in real estate and equipment, per E	Exhibit E		4,589,546.66
• • • • • • • • • • • • • • • • • • • •			
Total			\$49,159,571.05
Liabili	ITIES		
Current Funds:			
Accounts payable		\$2,591.34	
Reserve for valuation of books and accounts re-	ceivable	109,287.39	
Current Funds Surplus, per Exhibit C		945,686.29	\$1,057,565.02
Endowment and Other Special Funds:			
Due to Current Funds		\$218,647.00	
Principal of funds, per Exhibit D:		# 2 10,017.00	
Capital funds	\$40,146,472.16		
Special funds		43,293,812.37	43,512,459.37
,			
Plant Funds:			
Bequests, gifts, and income invested in plant			
Harriman Fund (see note, Exhibit D)		\$4 528 476 66	
		\$4,528,476.66 21,000.00	
		21,000.00	
Harkavy Fund		21,000.00 2,070.00	4,589.546.66
		21,000.00	4,589,546.66
Harkavy Fund		21,000.00 2,070.00 38,000.00	4,589,546.66

Ехнівіт В

STATEMENT OF INCOME AND EXPENDITURES FOR THE YEAR ENDED JUNE 30, 1952

For the Year Ended June 30, 1952		
INCOME:		
Investment income:		
Interest and dividends on securities	\$1,916,783.63	
Less amortization of bond premiums	28,214.05	\$1,888,569.58
Market value of stock dividend		15,577.87
Тотац		\$1,904,147.45
Less income allocated to Special Funds (Exhibit D)		167,458.45
<u> </u>		107,130.13
NET INVESTMENT INCOME APPROPRIATED FOR CURRENT		#4 #ac coo oo
PURPOSES (SCHEDULE 2)		\$1,736,689.00
Other income:		
Sales of publications	\$7,658.08	
Dormitory and mess hall	9,655.03	
Grants:	·	
American Cancer Society	11,400.00	
Carnegie Corporation of New York	23,000.00	
U. S. Public Health Service	24,770.17	
Miscellaneous		
TOTALLess amounts transferred to Special Funds (Exhibit D)	707.18	76,483.28
		70,403.20
TOTAL OPERATING INCOME (exclusive of net gain from		******
sales and redemptions of securities—see Exhibit D)		\$1,813,172.28
The state of the s		
Expenditures:		
Administration:		
Salaries		
Operating expenses		
Equipment	\$221,709.97	
Departmental Research Operations:		
Salaries		
Operating expenses		
Equipment		
Dormitory and mess hall:		
Salaries		
Operating expenses 5,987.26	1,350,777.72	
	1,000,777.72	
Office of Publications:		
Printing and publishing expenses \$23,185.64		
Office salaries* 10,984.83		
Stationery, postage, and other office expenses* 2,444.39	36,614.86	
Research Projects, Fellowships, Grants, etc.:		
Salaries		
Grants and miscellaneous		
Fellowship program:		
Grants and stipends		
Salaries	175,372.99	
Pension Fund—annuity and insurance	130 002 81	
Tension I and annuity and insurance		
Tomas proportions		1 004 270 27
Total expenditures		1,924,378.35
Total expenditures		1,924,378.35 \$111,206.07
Excess of expenditures over operating income—to Exhibit C.		\$111,206.07
Excess of expenditures over operating income—to Exhibit C.		\$111,206.07
		\$111,206.07
Excess of expenditures over operating income—to Exhibit C.		\$111,206.07
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses.	ditures for subsec	\$111,206.07
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses. Exhibit C Statement of Current Funds Surplut	ditures for subsec	\$111,206.07
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses.	ditures for subsec	\$111,206.07
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLY FOR THE YEAR ENDED JUNE 30, 1952	ditures for subsec	\$111,206.07 quent six months
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLY FOR THE YEAR ENDED JUNE 30, 1952	ditures for subsec	\$111,206.07 quent six months
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLU FOR THE YEAR ENDED JUNE 30, 1952 Balance, July 1, 1951	ditures for subsec	\$111,206.07 quent six months \$1,057,744.16
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLU FOR THE YEAR ENDED JUNE 30, 1952 Balance, July 1, 1951. Addition—Transfer from Special Funds (Exhibit D): Lecture Hall Fund.	ditures for subsec	\$111,206.07 quent six months \$1,057,744.16 1,050.00
*Represent expenditures for six months ended December 31, 1951; expension included in Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLU FOR THE YEAR ENDED JUNE 30, 1952 Balance, July 1, 1951. Addition—Transfer from Special Funds (Exhibit D): Lecture Hall Fund. Total.	ditures for subsec	\$111,206.07 quent six months \$1,057,744.16
*Represent expenditures for six months ended December 31, 1951; expensionly an Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLY FOR THE YEAR ENDED JUNE 30, 1952 Balance, July 1, 1951. Addition—Transfer from Special Funds (Exhibit D): Lecture Hall Fund. TOTAL Deductions:	ditures for subsec	\$111,206.07 quent six months \$1,057,744.16 1,050.00
*Represent expenditures for six months ended December 31, 1951; expensionly and in Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLY FOR THE YEAR ENDED JUNE 30, 1952 Balance, July 1, 1951. Addition—Transfer from Special Funds (Exhibit D): Lecture Hall Fund. TOTAL Deductions: Harriet H. Mayor Relief Fund (Exhibit D).	ditures for subsectors US \$1,901.80	\$111,206.07 quent six months \$1,057,744.16 1,050.00 \$1,058,794.16
*Represent expenditures for six months ended December 31, 1951; expensionly an Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLY FOR THE YEAR ENDED JUNE 30, 1952 Balance, July 1, 1951. Addition—Transfer from Special Funds (Exhibit D): Lecture Hall Fund. TOTAL Deductions:	ditures for subsectors US \$1,901.80	\$111,206.07 quent six months \$1,057,744.16 1,050.00
*Represent expenditures for six months ended December 31, 1951; expensionly and in Administration expenses. EXHIBIT C STATEMENT OF CURRENT FUNDS SURPLY FOR THE YEAR ENDED JUNE 30, 1952 Balance, July 1, 1951. Addition—Transfer from Special Funds (Exhibit D): Lecture Hall Fund. TOTAL Deductions: Harriet H. Mayor Relief Fund (Exhibit D).	\$1,901.80	\$111,206.07 quent six months \$1,057,744.16 1,050.00 \$1,058,794.16

	Balance June 30, 1952	\$32,000,000.00 7,856,555.84 103,310.80 2,828.80 171,609.72 10,888.42 1,278.58 10,157.69 2,824,447.80 3,830.45 577.42	62,944.49 188,674.39 30,000.00 17,337.97	\$43,293,812.37
Deductions	Expenditures or transfers	\$215,724.25 \$215,724.25 115.00 39,255.12 830.00	56,005.51	\$311,929.88
	Transfers to Current Funds Surplus (Exhibit C)		\$1,050.00	\$1,050.00
	Net realized gain on investments (Schedule 1)	\$950,852.61		\$950,852.61
	Transfer from Cur- rent Funds Surplus (Exhibit C)	\$1,901.80		\$1,901.80
Appitions	Income transferred from Cur- rent Funds (Exhibit B)	\$693.79		\$707.18
	Income from investments added to funds (Exhibit B)	\$424.97 150,451.10 155.62 123.52	15,577.87	\$167,458.45
	Gifts and sale of property	\$42,811.97	120,000.00	\$172,811.97
	Balance July 1, 1951	\$32,000,000.00 6,905,703.23 103,310.80 2,828.80 344,522.00 10,888.42 1,278.58 9,732.72 2,673,302.91 3,789.83 453.90 39,241.73 8,298.20	173,096.52 20,000.00 16,612.60	\$42,313,060.24
		Capital Funds: Endowment Fund. Capital Reserve Fund. Colburn Fund. Harkavy Fund. Teeple Fund. Van Gelder Fund. Special Funds: Bickel Fund. General Reserve Fund* General Reserve Fund* Harkavy Fund—income. Harriman Fund—income.	Lecture Hall Fund* Special Income Reserve Special Purpose Fund	TOTAL

*Outstanding contractual commitments for the building program at Cold Spring Harbor at June 30, 1952 aggregated approximately \$475,500, or \$241,000 in excess of the Harriman and Lecture Hall Funds available for this purpose. To cover the balance of the cost of completing the building program, the Trustees at their December 14, 1951 meeting appropriated an amount not to exceed \$350,000 from the General Reserve Fund.

EXHIBIT E

SUMMARY OF CHANGES IN INVESTMENT IN REAL ESTATE AND EQUIPMENT FOR THE YEAR ENDED JUNE 30, 1952

					CLASSI	CLASSIFICATION OF JUNE 30, 1952 BALANCE	30, 1952 BALANC	<u></u>
	Balance July 1, 1951	Additions	Deductions	Balance June 30, 1952	Buildings and grounds	Laboratory apparatus	Library	Operating equipment
DEPARTMENTS OF RESEARCH: Department of Plant Biology Stanford, California Department of Genetics*	\$158,957.20	\$815.63	: : : : :	\$159,772.83	\$73,229.67	\$42,237.16	\$25,117.33	\$19,188.67
Cold Spring Harbor, Long Island, New York	430,978.89	324,232.38	\$70,092.17	. 685,119.10	514,597.35	70,928.45	70,093.57	29,499.73
Washington, D. C	419,532.66	13,947.98	2,009.60	431,471.04	170,383.79	173,844.94	40,109.56	47,132.75
Cambridge, Massachusetts	21,348.20	302.63	521.60	21,129.23			1,193.94	19,935.29
Pasadena, California Department of Terrestrial Mag-	1,646,534.13	3,537.86		1,650,071.99	268,629.81	1,238,417.14	75,130.96	67,894.08
netism Washington, D. C	708,203.51	55,078.23†	106.00	763,175.74	401,418.69	248,311.77	40,449.60	72,995.68
Baltimore, Maryland	46,051.90	3,461.57		49,513.47		34,925.78	8,712.25	5,875.44
Total Departments of Research	\$3,431,606.49 \$401,376.28	\$401,376.28	\$72,729,37	\$3,760,253.40	\$1,428,259.31	\$1,808,665.24 \$260,807.21	\$260,807.21	\$262,521.64
Washington, D. C	823,366.79	8,347.05†	2,420.58	829,293.26	797,632.96			31,660.30
Total	\$4,254,973.28	\$409,723.33	\$75,149.95	\$4,589,546.66	\$2,225,892.27	\$1,808,665.24	\$260,807.21	\$294,181.94

^{*} See note on Exhibit D relative to construction of facilities for the Department of Genetics.

† Includes accumulated cost of equipment constructed and other adjustments at the Department of Terrestrial Magnetism amounting to \$39,828.32, and an adjustment at the Office of Administration amounting to \$1,255.00.

Schedule 1

SCHEDULE OF SECURITIES

Principal		Ma-	Book A	pproximate market
amount	Description	turity	value	value
	United States Government Bonds			
\$675,000	U. S. of America Treasury Bills	8-14-52	\$672,063.75	\$673,535
600,000	U. S. of America Treasury Ctf. of Ind. 178s	9-1-52	600,609.38	600,090
1,550,000	U. S. of America Treasury Ctf. of Ind. 17/8s	10-1-52	1,548,026.10	1,550,279
1,700,000	U. S. of America Treasury 2s	1954-52	1,700,000.00	1,699,468
3,550,000	U. S. of America Treasury 21/4s	1962-59	3,614,551.67*	3,520,046
1,000,000	U. S. of America Treasury 23/4s	1980-75	991,093.75	980,000
50,000	U. S. of America Savings Series "G" 21/2s	1953	50,000.00	49,300
50,000	U. S. of America Savings Series "G" 2½s	1954	50,000.00	49,100
50,000	U. S. of America Savings Series "G" 2½s	1954	50,000.00	48,950
100,000	U. S. of America Savings Series "G" 2½s	1955	100,000.00	97,600
100,000	U. S. of America Savings Series "G" 2½s	1956	100,000.00	97,000
100,000	U. S. of America Savings Series "G" 2½s	1957	100,000.00	96,400
100,000	U. S. of America Savings Series "G" 2½s	1958	100,000.00	95,800
100,000	U. S. of America Savings Series "G" 2½s	1959	100,000.00	94,900
100,000				
	U. S. of America Savings Series "G" 2½s	1960	100,000.00	94,800
\$9,825,000	Total U. S. Government		\$9,876,344.65 	\$9,747,268
	FOREIGN AND INTERNATIONAL BANK BONDS			
\$300,000	Aluminum Company of Canada, Ltd., S. F. Deb. 31/8	1970	\$304,500.00	\$308,250
100,000	Australia, Commonwealth of, S. F. 31/4s	1956	100,000.00	95,125
50,000	Australia, Commonwealth of, S. F. 31/4s	1957	48,750.00	46,500
100,000	Canadian National Ry. Co., 4½s Guar	1957	112,000.00	108,625
125,000	International Bank for Reconstruction and Development, 3s	1976	125,000.00	117,812
125,000	International Bank for Reconstruction and Development, 33/8s	1975	123,125.00	124,218
200,000	Shawinigan Water & Power Co., 1st Mtg. & Coll. Tr. S. F. 3s			
	Series "M"	1971	206,840.00*	180,000
\$1,000,000	Total Foreign and International Bank		\$1,020,215.00	\$980,530
-				
	Public Utility Bonds			
\$300,000	American Gas & Electric Co., Serial Note 2½s	1953-55	\$304,103.83*	\$298,375
125,000			128,141.63*	
	Columbia Gas System, Inc., Deb. 3s.	1975		120,625
237,000	Columbus & Southern Ohio Electric Co., 1st Mtg. 3¼s	1970	248,597.02*	241,740
300,000	Consolidated Natural Gas Co., Deb. 23/4s	1968	300,693.82*	297,000
200,000	Minnesota Power & Light Co., 1st Mtg. 31/8s	1975	203,828.27*	193,000
200,000	Northern Natural Gas Co., Serial Deb. 25/8s	1954-55	203,186.92*	198,875
100,000	Ohio Power Co., 1st Mtg. 3¼s	1968	101,500.00	104,000
200,000	Panhandle Eastern Pipe Line Co., Serial Deb. 23/4s	1961-62	201,818.01*	193,125
100,000	Panhandle Eastern Pipe Line Co., S. F. Deb. 31/4s	1973	101,521.22*	99,000
50,000	Philadelphia Electric Co., 1st & Ref. Mtg. 21/8s	1978	49,687.50	49,062
207,000	Philadelphia Electric Power Co., 1st Mtg. 25/8s	1975	211,025.62*	194,580
200,000	Public Service Co. of Indiana, Inc., 1st Mtg. 31/8s	1975	203,864.77*	200,500
45,000	Puget Sound Power & Light Co., 1st Mtg. 41/4s	1972	46,278.85*	47,250
210,000	Tennessee Gas & Transmission Co., 1st Mtg. Pipe Line 23/4s	1966	212,205.00*	199,500
191,000	Tennessee Gas Transmission Co., 1st Mtg. Pipe Line 3s	1969	195,949.78*	183,360
100,000	Tennessee Gas Transmission Co., S. F. Deb. 41/8s	1971	102,200.44*	106,750
265,000	United Gas Corp., 1st Mtg. & Coll. Tr. 23/4s	1967	265,000.00	249,100
\$3,030,000	Total Public Utility		\$3,079,602.68	\$2,975,842
				=======================================

^{*} After deduction for amortization of premiums on bonds purchased subsequent to January 1, 1940.

SCHEDULE 1 SCHEDULE OF SECURITIES—Continued

			,	Approximate
Principal		Ma-	Book	market
amount	Description	turity	value	value
	Communication Bonds			
\$150,000	American Telephone & Telegraph Co., Conv. Deb. 23/4s	1961	\$153,680.07*	\$163,312
		1975		
150,000	American Telephone & Telegraph Co., Deb. 23/4s		152,081.25*	141,750
200,000	American Telephone & Telegraph Co., Conv. Deb. 33/8s	1963	223,460.50*	232,000
200,000	Mountain States Telephone & Telegraph Co., Deb. 31/88	1978	201,470.00*	200,000
100,000	New York Telephone Co., Ref. Mtg. 31/88	1978	101,232.76*	100,500
200,000	Pacific Telephone & Telegraph Co., Deb. 31/4s	1978	204,635.44*	204,250
300,000	Southwestern Bell Telephone Co., Deb. 31/8s	1983	306,500.00*	301,500
\$1,300,000	Total Communication		\$1,343,060.02	\$1,343,312
	RAILROAD BONDS			
\$100,000	Chesapeake & Ohio Ry. Co., Gen. Mtg. 4½s	1992	\$99,464.29	\$120,000
300,000	Fort Worth & Denver Rwy. Co., 1st Mtg. 43/8s	1982	303,000.00	308,625
			· ·	
100,000	Pennsylvania R. R. Co., Cons. Mtg. 4½s	1960	104,662.50	108,500
\$500,000	Total Railroad		\$507,126.79	\$537,125
	RAILROAD EQUIPMENT TRUST BONDS			
M470 000		1057 50	0146 240 21	0444.645
\$150,000	Chesapeake & Ohio Ry. Co., Eq. Tr. 2s Guar	1956-58	\$146,340.34	\$144,610
300,000	Chicago Burlington & Quincy R. R. Co., Eq. Tr. 21/4s Guar	1958-63	292,507.12	283,435
100,000	Great Northern Railway Co., Eq. Tr. 2s Guar	1960–61	98,538.91	92,535
150,000	Pennsylvania R. R. Co., Eq. Tr. 23/8s Guar	1958–62	146,358.96	142,401
100,000	Southern Pacific Co., Eq. Tr. 21/8s Guar	1956 & 59	100,135.53*	95,775
150,000	Southern Pacific Co., Eq. Tr. 21/8s Guar	1956-58	146,251.10	144,915
150,000	Southern Railway Co., Eq. Tr. 21/8s Guar	1956-58	145,928.69	144,280
\$1,100,000	Total Railroad Equipment Trust		\$1,076,060.65	\$1,047,951
	Industrial and Miscellaneous Bonds			
\$200,000	Aluminum Company of America, S. F. 31/8s	1964	\$200,000.00	\$204,750
187,000	American Tobacco Co., Deb. 3s	1969	189,390.33*	188,870
234,000	Bristol Myers Co., Deb. 3s	1968	234,931.86*	234,000
300,000	C. I. T. Financial Corp., Deb. 25/8s	1959	301,031.15*	294,000
150,000	Dow Chemical Co., Deb. 2.35s	1961	150,579.55*	144,750
153,000	Food Machinery Corp., S. F. Deb. 21/28	1962	152,308.98	147,262
275,000	Goodrich (B. F.) Company, 1st Mtg. 23/4s	1965	275,868.45*	272,250
267,000	P. Lorillard Co., Deb. 3s.	1963	273,304.51*	270,337
295,000	National Dairy Products Corp., Deb. 23/4s	1970	298,696.46*	288,731
488,000	Phillips Petroleum Co., S. F. Deb. 23/4s	1964	493,378.71*	488,610
125,000	Pittsburgh Plate Glass Co., S. F. Deb. 3s	1967	125,000.00	127,343
150,000	Quaker Oats Co., Deb. 25/8s	1964	148,922.50	147,375
300,000	Seagram (Joseph E.) & Sons, Inc., Deb. 2½s	1966	298,500.00	279,000
300,000	Service Pipe Line Co., S. F. Deb. 3.20s.	1982	300,000.00	301,500
500,000	Shell Union Oil Corp., Deb. 2½s	1982	503,895.35*	473,125
500,000	Socony-Vacuum Oil Co., Inc., Deb. 2½s	1976	489,528.75	468,125
300,000	Swift & Co., Deb. 25/8s	1972	301,426.65*	290,250
500,000	Texas Corporation, Deb. 3s.	1965	521,431.43*	510,000
346,000	Union Oil Company of California, Deb. 23/4s	1970	355,896.07*	337,350
57,000	Union Oil Company of California, Conv. Sub. Deb. 31/8s	1972	57,000.00	61,488
400,000	Westinghouse Electric Corporation, Deb. 25%s	1971	403,798.52*	384,000
\$6,027,000	Total Industrial and Miscellaneous		\$6,074,889.27	\$5,913,116
\$22,782,000	Bonds—Funds Invested		\$22,977,299.06	\$22,545,144
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^{*} After deduction for amortization of premiums on bonds purchased subsequent to January 1, 1940.

			pproximate
Number of shares	Description	Book value	market value
onares	Preferred Stocks	, a. a.	, and
		*	
	Air Reduction Company, Inc., 4.50% Cum. Conv. Pref	\$41,195.04	\$42,900
	Anchor Hocking Glass Corp., \$4.00 Cum. Pref	112,750.00	102,500
	Appalachian Electric Power Co., 4½% Cum. Pref	159,000.00	157,875
	Armstrong Cork Co., \$3.75 Cum. Pref	205,500.00	194,500
1,500	Bethlehem Steel Corp., 7% Cum. Pref	183,637.50	210,000
500	Case (J. I.) Co., 7% Cum. Pref	62,225.00	76,000
1,500	Celanese Corp. of America, 4½% Cum. Conv. Pref. "A"	152,041.50	154,500
600	Cleveland Electric Illuminating Co., \$4.50 Cum. Pref	68,112.25	64,575
1,900	Consolidated Edison Co. of N. Y., Inc., \$5.00 Cum. Pref	202,815.50	203,775
1,125	Continental Can Co., Inc., \$3.75 Cum. Pref	115,312.50	106,312
1,000	Continental Can Co., Inc., \$4.25 Cum. Conv. 2nd Pref	106,779.99	114,000
600	Corn Products Refining Co., 7% Cum. Pref	110,335.18	105,600
2,075	duPont (E. I.) de Nemours & Co., \$4.50 Cum. Pref	235,401.89	245,368
1,000	El Paso Natural Gas Co., 4.10% Cum. Pref	111,442.21	87,000
2,000	General Foods Corp., \$3.50 Cum. Pref	201,000.00	190,000
1,500	General Motors Corp., \$5.00 Cum. Pref	187,937.50	185,812
1,000	General Shoe Corporation, \$3.50 Cum. Pref	102,250.00	88,000
1,000	Grant (W. T.) Co., $3\frac{3}{4}\%$ Cum. Pref	100,447.91	95,500
300	Merck & Co., Inc., \$4.00 Conv. Cum. 2nd Pref	31,200.00	33,000
800	National Distillers Products Corp., 4¼% Cum. Conv. Pref	80,000.00	80,200
2,000	Niagara Mohawk Power Corp., 3.60% Cum. Pref	207,990.00	175,000
1,300	Ohio Power Co., 4½% Cum. Pref	144,630.02	139,425
1,500	Pacific Telephone and Telegraph Co., 6% Cum. Pref	235,220.75	213,000
1,000	Panhandle Eastern Pipe Line Co., 4% Cum. Pref	104,166.68	97,750
1,000	Pillsbury Mills, Inc., \$4.00 Cum. Pref	107,722.00	100,000
2,000	Reynolds (R. J.) Tobacco Co., 3.60% Cum. Pref	199,683.75	173,000
784	Sherwin-Williams Co., 4% Cum. Pref	86,410.03	82,516
1,300	Standard Oil Co. of Ohio, 33/4% Cum. Pref. "A"	139,976.28	126,750
3,100	U. S. Steel Corp., 7% Cum. Pref	443,407.57	432,450
37,284 	Total Preferred Stocks	\$4,238,591.05	\$4,077,308
	Common Stocks		
2,000	Allied Chemical & Dye Corp	\$96,175.97	\$149,750
2,400	Aluminium Limited	204,826.34	247,500
19,360	American Can Company	445,351.97	663,080
1,300	American Cyanamid Co	161,924.48	149,500
6,534.15	American Gas and Electric Company	280,046.46	385,514
2,850	American Telephone & Telegraph Co	413,951.54	439,612
	2,850 rights to subscribe for American Telephone & Telegraph Co., 3½% Conv. Deb. due 1964		6,234
3,000	Anderson, Clayton & Co	164,768.84	139,500
4,700	Armstrong Cork Company	231,516.80	235,000
2,800	Atchison, Topeka and Santa Fe Rwy. Co.	232,758.69	255,500
3,000	C. I. T. Financial Corporation.	151,369.19	193,875
5,200	Caterpillar Tractor Co	272,536.55	274,300
2,000	Chase National Bank of the City of New York.	71,361.04	76,750
60	Christiana Securities Co.	356,143.00	340,800
3,200	Chrysler Corporation.	145,078.62	250,800
5,400	Cleveland Electric Illuminating Company.	227,773.54	279,450
2,000	Consolidated Edison Company of N. Y., Inc.		69,500
2,750	Consumers Power Co	94,687.50	97,625
1,250	Continental Illinois National Bank & Trust Co. of Chicago	88,175.00	104,375
6,373	Continental Insurance Co	241,723.37	487,534
13,500	Continental Oil Co. of Delaware.	265,129.64	909,562
1,000	Corning Glass Works	59,631.83	74,000
1,000	Coming Ciaco Works	37,031.03	74,000

(Continued on following page)

SCHEDULE OF SECURITIES—Continued

SCHEDULE 1

Number of		Book	Approximate market
shares	Description	value	value
	COMMON STOCKS—Continued		
6,100	Delaware Power & Light Company	\$128,803.87	\$145,637
5,300	duPont (E. I.) de Nemours & Co	216,311.84	459,775
9,900	Eastman Kodak Co	272,756.19	430,650
2,500	Fidelity-Phenix Fire Insurance Co. of New York	171,032.77	195,000
3,620	Fireman's Fund Insurance Co	128,858.78	208,150
11,700	General Electric Co.	461,324.90	735,637
4,000	General Motors Corporation	167,302.84	185,000
7,800	General Motors Corporation	207,915.99 85,843.58	455,325 156,200
2,200 400	Guaranty Trust Co. of N. Y.	98,003.91	118,000
13,800	Gulf Oil Corp.	310,532.07	769,350
2,900	Gulf States Utilities Co.	61,802.82	70,325
1,800	Halliburton Oil Well Cementing Co.	79,847.34	95,850
2,567	Hartford Fire Insurance Co.	173,120.24	370,931
13,600	Humble Oil & Refining Co	226,988.35	1,047,200
2,500	Illinois Power Co	97,697.35	93,125
2,800	Inland Steel Company	156,896.54	132,300
7,940	Insurance Company of North America	243,482.20	674,900
3,305.25	International Business Machines Corp	213,004.45	723,849
5,000	International Nickel Co. of Canada, Ltd	185,533.15	226,250
1,800	International Paper Company	95,379.27	85,275
3,900	Johns-Manville Corp	144,315.72	291,525
9,000	Kennecott Copper Corporation	459,524.19	700,875
4,000	Kimberly-Clark Corporation	178,079.45	170,500
7,000	Kresge (S. S.) Company	220,699.69	251,125
320	Mellon National Bank and Trust Company	67,193.07	108,800
8,000	Mercantile Stores Company, Inc	174,200.45	136,000
9,000	Merck & Co., Inc.	172,889.46	245,250
7,000	Middle South Utilities, Inc	168,660.65 134,007.13	162,750
5,800	Monsanto Chemical Co	244,668.33	311,750 553,175
5,800 1,300	Montgomery Ward & Co., Inc.	70,154.45	83,850
7,400	National Cash Register Co	257,367.19	422,725
2,200	National City Bank of New York.	88,087.50	101,475
10,000	Newberry (J. J.) Co	132,451.57	398,750
8,000	North American Co	172,640.38	179,000
3,300	Ohio Edison Co	105,150.00	113,437
7,000	Penney (J. C.) Co	248,413.74	490,000
1,000	Peoples Gas Light and Coke Company	106,350.00	137,000
6,700	Pfizer (Chas.) & Co., Inc.	138,192.65	237,850
2,400	Phelps Dodge Corporation	71,057.69	84,600
3,990	Philip Morris & Co., Ltd., Inc	210,697.55	184,537
8,800	Pittsburgh Plate Glass Co	283,938.09	427,900
4,200	Procter & Gamble Co	177,227.28	270,900
2,000	Public Service Co. of Indiana, Inc.	57,045.34	62,250
4,600	Scott Paper Co.	103,055.95	246,100
2,600	Seaboard Oil Co. of Delaware	229,104.22	228,150
15,200 500	Sears, Roebuck & Co	347,030.43 35,039.88	828,400 41,500
		50,819.74	56,875
1,300 4,800	Sharp & Dohme, Inc	294,227.78	324,000
9,000	Socony-Vacuum Oil Co., Inc.	272,564.13	352,125
5,000	Southern California Edison Company.	175,996.33	175,000
8,700	Standard Oil Co. of Indiana	315,458.41	740,587
7,827	Standard Oil Co. of New Jersey	241,389.50	633,987
6,200	Texas Company	163,636.60	366,575
8,000	Union Carbide & Carbon Corp	250,787.83	540,000
5,000	United Fruit Company	141,876.60	315,000
8,050	United Gas Corp	127,987.59	219,362

(Concluded on following page)

Schedule	1 Schedule of Secu	URITIES—C	oncluded		
Number of shares	Description			Book value	Approximate market value
	Common Stocks—Con	cluded			
4,700 4,750 4,000 4,700 10,100 2,500 447,846.4 485,130.4	United States Gypsum Co United States Plywood Corporation United States Steel Corporation Virginia Electric and Power Co Westinghouse Electric Corp Weyerhaeuser Timber Company Total Common Stocks COMMON AND PREFERRED STOCKS— AGGREGATE INVESTMENTS (BONDS A	-Funds Invest	ed.	. 174,129.76 . 188,577.48 . 92,038.13 . 276,109.53 . 130,813.92 . \$16,174,393.61 . \$20,412,984.66	143,687 160,000 111,625 395,162 170,625 \$25,923,774 \$30,001,082
July 1, 1951-	SUMMARY OF SECURITY TRANSACTI —Cash awaiting investment	•		,	\$109,269.04
		Gain	Loss	Book value	
Preferred sto Common sto	cks. cks. rights.	\$17,110.81 929,278.79 4,567.35	\$104.34	\$7,900,474.13 2,204.34 1,850,463.73	
Net Gain—t	o Exhibit D	\$950,956.95	\$104.34 950,852.61	\$9,753,142.20 950,852.61	10,703,994.81
			\$950,956.95		
	ied to amortization of bond premiumse of stock dividend				28,214.05 15,577.87
Cash transfe	rred for current needs				\$10,857,055.77 165,000.00
				;	\$10,692,055.77
	Асоци	SITIONS			
Preferred sto	cks,			\$8,541,116.65 259,175.03 1,769,588.44	10,569,880.12
June 30, 195	2—Cash awaiting investment				\$122,175.65
,				=	

SCHEDULE 2

SUMMARY OF CHANGES IN CURRENT FUNDS SURPLUS ACCOUNTS FOR THE YEAR ENDED JUNE 30, 1952

	Balance June 30, 1952		\$9,915.82	1,125.47	175.10	5,312.56	20,976.09	4,010.99	23 020 36	5,235.36	206.50	4,747.78	\$100,208.23	2,696.65 28,754.13	52,857.21	128,479.80	02 020 04	150,619.68	\$945,686.29	
Deductions	Allotments and transfers		\$10,054.46	2.181.62			12,633.69	14,646.13	17.293.54	4,871.59			\$80,536.46	2,263.13 104,449.34	5,979.08	65,000.00	41 208 90		\$299,536.81	
DEDU	Expendi- tures		\$101,383.56	176.465.68	7,865.73	16,266.11	244,549.93	95,410.80	325,982.22	106,065,44	5,497.75	5,582.45	\$1,350,777.72	221,709.97	23,185.64	175,372.99		139,902.81	\$1,924,378.35	
	Grants, other credits, sales of publications, etc.			\$79.79	7,400.00	17,840.42 9,655.03		57.87	17:00		4,000.00	6,929.75	\$46,121.07		7,658.08	23,000.00	•		\$76,779.15	
SNOI	Transferred from Special Funds (Exhibit D)										:						\$1.050.00		\$1,050.00	
ADDITIONS	Allotments and transfers		\$6,474.23	13,837.00			42,680.14	2,300.00	47,049.16	5,255.00			\$125,595.53	40,586.36 1,250.02	550.00	15,628.00	107 427 72	6,301.51	\$297,339.14	
	Trustees' appropriations		\$98,950.00	168,577.00		1,200.00	217,665.00	101,350.00 271.300.00	296,043.00	107,654.00			\$1,262,739.00	184,750.00	18,450.00	47,500.00		125,000.00	\$1,736,689.00	
	Balance July 1, 1951		\$15,929.61	1,123.47 6,821.40	640.83	857.88	17,814.57	10,360.05	23,203.96	3,263.39	1,704.25	3,400.48	\$97,066.81	1,333.39	48,384.77	61.471,787	414.901.67	159,220.98	\$1,057,744.16	
		Reserves for Unexpended Appropriations: Departmental Research Operations:	Department of Plant BiologyResearch Cornoration—orant	Department of Genetics	American Cancer Society—grant	Dormitory and mess hall	Geophysical Laboratory.	Department of Archaeology	Department of Terrestrial Magnetism	Department of Embryology	II & Dublic Hoolth Commission	O. S. I ubile meanin Selvice—grant		Administration General Operations.	Office of Publications Research Projects Editorical Courts	According 1 tolectes, regionships, Grants, etc	General Contingent Fund.	Pension Fund.	TOTAL	



REPORT OF THE PRESIDENT

OF THE

CARNEGIE INSTITUTION OF WASHINGTON

FOR THE YEAR ENDING JUNE 30, 1952



REPORT OF THE PRESIDENT

OF THE

CARNEGIE INSTITUTION OF WASHINGTON

It is now fifty years since the Carnegie Institution of Washington was founded; and as we look back on the program of research begun in 1902 we can see that it has had a far-reaching influence. We may legitimately feel that the Institution has proceeded in the way that Andrew Carnegie intended and that the policies that have been followed would receive his approbation. The aim prescribed for us has been to "encourage, in the broadest and most liberal manner, investigation, research, and discovery, and the application of knowledge to the improvement of mankind." The means we have chosen for the achievement of this aim has been to carry on programs of fundamental research in our own continually evolving departments and to support scholars working independently on related programs.

The difference between fundamental and applied research must be kept constantly in mind. It is a difference that lies not so much in methods and procedures as in motivation and objectives. Scientists conduct their investigations in much the same manner whether they are employed by a disinterested institution such as ours, by one of the agencies of the federal government, or by a large industrial corporation. They differ only in what they are trying to accomplish. The controlling purpose of fundamental research is to add piece by piece to our knowledge of the universe we live in and of ourselves. It is essentially, as the Greeks regarded it, a part of the larger field of knowledge and thought known as philosophy. It may and often does yield, or at least lay the foundations for, important practical applications, but these are incidental to its main purpose. Sometimes a more direct contribution to the practical application of fundamental research occurs when a scientist who has worked in a special field for years temporarily lends his talents to a program of development, as was done by many staff members of the Institution during the first and second World Wars. On the other hand, the purpose of applied science is more definite and immediate. Some of the larger corporations maintain impressive laboratories and able scientific staffs to whom they give great freedom of action. Their primary objective is to bring about the discovery of new processes or new products that will sooner or later—not too much later—yield a profit on the money invested. If the gain redounds to others beside the corporations or to the whole of society, as it often does, this circumstance does not alter the fact that the reason of the corporations for maintaining research establishments is primarily to improve their products and performance, and thus render their undertakings more successful for their stockholders. The greater part of the research conducted under the immediate sponsorship of the government is similarly directed toward definitely anticipating practical ends—the protection of the government as a consumer, the development of more effective weapons of defense, the improvement of agricultural methods, the development of cheaper or more abundant sources of power, and so on. The developmental or applied research of both industry and government may and often does contribute significantly to our knowledge and understanding of the universe, but its contribution is incidental to its main purpose.

The Institution has always devoted its

main resources to fundamental research, and it has no thought of changing its policy. This is as it should be. Americans have a natural facility for applying science effectively to the improvement of their daily lives without the stimulus of any form of subsidy. They have not always in the past shown an equal appreciation of the underlying importance of basic science for all practical research efforts. Ours has been a pioneering country pioneering first in the geographical sense and pioneering more recently in intricate new modes of living-and the active pioneer seldom has time to contemplate the philosophical meanings of his existence or to develop the subtleties of color and form. Fortunately there is a growing realization that basic and applied science are two important parts of a single endeavor, and as a result the United States is now forging ahead in the field of fundamental research. An important aspect of this awakening realization is the entrance of government into the support of basic science to a significant degree.

The Institution's devotion to fundamental research has simplified many of our problems and our relations with persons and interests outside our immediate circle. We have, for instance, no great problem in our relations with industry. Our laboratories are wide open, and competent, serious-minded visitors are always welcome. We promptly publish what we learn for the benefit of all. When our researches have practical implications, these implications are usually remote, and for that reason there is no scramble for first news of a discovery. In this respect we differ from laboratories where applied research is being carried on. In such laboratories, especially in engineering schools, there is need for very close contact between the individual research worker and industry in order that the current work may have reality and vitality. This contact often takes the form of consultation service rendered to an interested industrial establishment in the neighborhood. The relation can be healthy when it is conducted with reasonableness. In our Institution, however, there is no need for formalized industrial contact; and although our staffs often participate in activities outside their departments-lecturing at neighboring universities or attending scientific gatherings that are of interest to them—our relations with industry are entirely on an informal rather than an individual or contractual basis.

In handling problems that relate to patents on inventions, the Institution has followed a policy which is generally regarded as sound and is adhered to by the entire staff. Patent matters are not a subject of controversy with us, or even a matter of moment in our scheme of operations. When an invention occurs, a patent is applied for if this appears to be in the public interest. We do not forget that the intent of the founding fathers in providing in the Constitution the basis for patent legislation was to further the introduction of novel devices for the common good by protecting the inventor or his assignees against early and destructive competition. They also contemplated a grant to an inventor in return for the full disclosure of his ideas, to avoid the kind of industrial secrecy which was the practice before patent systems appeared. This country has prospered greatly under its system of free enterprise, and no small part of its extraordinary progressiveness in practical ways has been due to the existence, through its period of rapid expansion, of a workable patent system. The system may need to be modernized, but the principles which governed those who established it still hold good; there are valuable inventions which would never have come into use if not thus protected. And in this spirit the Institution deals with patent matters when it considers it essential to do so. But most of the inventions made in the Institution are of such a nature that they will automatically come into use if they are of real merit, and therefore they need no legal protection. Often they are merely parts of the paraphernalia of research, which can be readily adopted and improved upon by others; and in such cases publication alone suffices. Although it is not generally realized, experimental scientists habitually and frequently make inventions as they proceed—patentable ones, for that matter, but usually limited in their application to the specific problem in hand.

There is no sound reason why the Institution should not under proper conditions derive an income from an invention produced in its laboratories and use that income to further research for the public benefit. It has never done so, to be sure, and the occasion for doing so may not arise. The point is that whatever occurs in the laboratories of the Institution should be treated in such a manner as to result in the greatest possible public benefit, and the means to that end should be examined critically in each particular case. But it is important to remember that this is of minor concern to us; for nearly all our work produces its effects on thinking rather than on action. If in the long run our work has practical implications, we shall usually have performed our full duty if we are diligent in making our results generally known as promptly as possible.

From the point of view of the nation as a whole, the bridge from fundamental discovery to practical application is not always and everywhere effective. Much of the fundamental work is done by universi-

ties, research institutions such as our own, and government; and all these are lame in moving forward to applications when they try to do so. It takes the resourcefulness and drive of competitive industrial effort to vault the many hurdles that are always present when novel methods or devices are introduced into the current scene. To introduce a new method or device requires the expenditure of venture capital, which is not available in the ordinary sense to public or semipublic institutions. There is nothing wrong about a fundamental idea's flowing out of laboratories such as ours and later becoming a matter of vigorous industrial exploitation if it is all done within the framework of the law and with proper regard for the public interest. We believe in private enterprise in this country even though some of our citizens often give the contrary impression. But in our system of private enterprise it is not entirely clear where the burden of the initiative lies for bringing a promising idea to the point where it will be exploited. And consequently there is often a long wait before industry on its own initiative takes effective action. We have, in a word, no effective organization in the country as a whole for bringing the fruits of disinterested research to the point where they invite industrial development. It was the existence of this gap in organization which led to the creation of Research Corporation by Frederick G. Cottrell and his associates, but the gap remains wide in spite of their efforts. This does not mean that the Institution ought to plunge into the application of science. It does mean that in order to ensure prompt public benefit from its findings, the Institution sometimes needs to look ahead beyond its immediate program of research and publication.

An interesting case in point developed very early in the Institution in connection

with hybrid corn. The most important fundamental work on this subject was carried out by George H. Shull at the Institution's laboratory at Cold Spring Harbor and was duly published. He painstakingly developed, through the process of self-fertilization, scrawny and relatively unattractive lines of corn; and he demonstrated beyond question the hybrid vigor that then arose from crosses. All this became known, but known only in scientific circles; and practical men were far away and thinking of other things. A decade elapsed before there was real action. Then under vigorous industrial development, with the use of double crossing methods, the whole growing of corn in this country was revolutionized. Today it is estimated that the use of hybrid corn, as compared with the older methods, represents an economic advantage in this country alone of some three billion dollars a year. The question naturally arises whether some action on the part of the Institution itself could have shortened that decade of inaction.

We have the problem before us now in connection with the mass cultivation of algae. From a program of research on photosynthesis at our Department of Plant Biology it appeared a few years ago that it was possible to grow the green alga Chlorella in such manner that it would produce 56 per cent dry weight of protein, and this was news of importance in a world where there are great protein-starved populations. This time the Institution proceeded somewhat farther than it has on previous occasions; for while it devoted its own energies almost entirely to the fundamental aspects of the subject, it did collaborate with other institutions in exploring the practical aspects. In fact, with the aid of Research Corporation it collaborated with Stanford Research Institute in an exploratory program, and then, with the aid

of Carnegie Corporation of New York, it called in a firm of consulting engineers, Arthur D. Little, Inc., which built a pilot plant and studied the economics of the situation. All this has resulted in the preparation of a monograph, covering our own work and that of others, which summarizes the status of this whole development as a possible means of increasing the world's critical food supply. It is highly improbable even now that the exact methods and the particular organism utilized by the Institution and its collaborators will ultimately prove to be the most successful ones. The whole project is but one phase of a much larger one: the production of food by process methods rather than by conventional agriculture. There is no doubt that in the future man will produce a very significant part of his food supplies by using the methods and some of the apparatus of the chemical engineer, combined with the biologist's growing knowledge of the various organisms, the way in which they thrive, and the products which can come from them. The effort of the Institution has been devoted to bringing closer the time when this could practically be accomplished.

In short, I believe that the Institution's primary responsibility is to carry on fundamental research in science with no initial view to its industrial application; but when in the course of its fundamental research ideas and principles evolve that promise material advantage to society, it is proper for the Institution to carry its studies beyond the clarification of principles and explore some of the broader possibilities of use for human betterment. We can follow this course remembering that Andrew Carnegie in his letter to the trustees stated as one of the principal objects of the Institution "the application of knowledge to the improvement of mankind." We may

ourselves, in important instances, enter the field of development momentarily, or collaborate with others as they do so, without losing sight of primary responsibilities, since our entire staff is fully in accord with the Institution's policy of devoting its main effort to increasing man's fundamental knowledge.

Looking back over the history of the Institution we see many ways in which the research which we conducted at the time because of our basic interest in contributing to an understanding of the cosmos, the earth, and the creatures that inhabit the earth has later proved to be basic to important undertakings or practical applications. I shall mention only a few.

Much has been written about the origins of radar: that extraordinary technique which determined the outcome of the Battle of Britain by enabling relatively few fighters to combat great waves of attacking bombers. This was the occasion of Winston Churchill's classic remark that "never in the field of human conflict was so much owed by so many to so few." This is not the place to treat the history of the whole subject; indeed, in view of some of the recent controversial claims, the subject is not altogether a pleasant one. The important part is that during the war British and American scientists worked together effectively and in harmony and produced a host of devices which affected the course of the war favorably and in many ways. Nor is it necessary here to develop the full history of the early origins of radar. One fact stands out. Radar, in its essence, is the method of locating objects in space by propagating a beam of short pulses of electromagnetic energy and measuring the time between the pulse and its echo at the sending station. The pulsation is an essential element of the process. On July 10, 1925, Merle A. Tuve and Gregory Breit, then both of the Department of Terrestrial Magnetism, sent out pulses from a Navy transmitter they had modified for the purpose and observed echoes from the ionosphere. During the next few years, as they were studying the ionosphere, they were troubled by echoes coming from airplanes, which interfered with their measurements. There is no doubt whatever that these experiments introduced the first use of short-wave electromagnetic pulses to observe anything whatever, the first effective electrical method of surveying the ionosphere, and the first observation of the echoes of pulses from airplanes. This was not the first detection of objects by electrical means, but it was the first radar. The fact that after its successful appearance it was devoted to objectives in fundamental science was fully in accordance with the traditions of the Institution. The methods and results were promptly published. In ionospheric research they were generally adopted. They were also developed, by the efforts of many men in several countries, into practical devices for aircraft detection, the navigation of ships, and many other purposes.

Out of the Geophysical Laboratory there appeared early discoveries of far-reaching practical significance for the manufacture of various kinds of glass. As early as 1906 Arthur L. Day in collaboration with E. S. Shepherd published a pioneering paper on low-expansion quartz glass. Day's conception of the possibilities inherent in the use of high percentages of silica, and his early experiments, were basic to the development of Pyrex and similar glasses.

As a result of his experience thus acquired in the field of glass chemistry, Day was selected by the War Industries Board in World War I to take charge of the development of optical-glass production in the United States. Under his direction

members of the Laboratory were given full authority to prescribe the materials and techniques used in the various glass-producing plants to which they were severally assigned. This program not only supplied the government with 97 per cent of the total quantity of optical glass needed by it for war purposes, but introduced a new method for its manufacture and resulted in the permanent establishment of a new industry in the United States. The Laboratory's basic study of the soda—lime—silica system, which for the first time gave a scientific understanding of the five-thousand-year-old glass industry, resulted in an enormous spurt in research by the industry.

After the war the Laboratory continued active research for the improvement of annealing methods, and in 1920 produced as one of its Geophysical Laboratory Papers the classic work on the subject by Leason H. Adams and E. D. Williamson. Later, in 1935, a modified formula for the annealing of Pyrex glass in large castings was published, which proved to be fundamental to the successful manufacture of the 200-inch Pyrex mirror for the Hale telescope. The invention by George W. Morey, now Acting Director of the Laboratory, of lanthanum and thorium borate glasses with high refractive index and low dispersion led to the introduction of a new family of glasses of great importance in the design of photographic lenses.

The Geophysical Laboratory also contributed in an important way to the development of the Portland cement industry. In 1915 one of its staff members, G. A. Rankin, published a paper entitled "The ternary system CaO—Al₂O₃—SiO₂," with an optical study by Fred. E. Wright. This paper solved scientifically the age-old riddle of cement, and it has served ever since as a guide for the chemical phases of the cement industry. Rankin made a

systematic study of each constituent, separately and in combination with the others, and determined with assurance the structure of cement and its behavior. His investigations were the more remarkable because they were carried on in a most painstaking and laborious manner before the development of certain time- and laborsaving laboratory equipment that is now available. Rankin's work made it possible to predict the behavior of any mixture of lime, alumina, and silica so that the best composition of cement for a given purpose can be determined before the product is tried out.

A practical by-product of research at the Department of Embryology, developed incidentally to facilitate its own experiments, was an instrument devised by S. R. M. Reynolds and constructed with the aid of Osborne O. Heard called a tokodynamometer, for recording the contractions of the uterus when a child is born. By the use of ingenious devices employing strain gauges, which are highly sensitive to local pressure and which can be applied to the patient's abdomen, the contractions of the underlying uterus may be detected at several places at once and may be recorded on a paper tape. The apparatus is useful in studies of normal and abnormal births, and was put into use at once in the Woman's Clinic of Johns Hopkins Hospital in collaboration with members of the Department of Obstetrics of Johns Hopkins University. The instrument is being manufactured by Statham Laboratories, Inc., of Los Angeles, and about sixty have been put into use.

The information about human embryology gathered at the Department and published in "Contributions to Embryology" forms a conspicuous element in every textbook on the subject published in the past decade. Motion pictures of developing em-

bryos and of cellular biology done under the direction of Warren H. Lewis are being used widely by colleges and professional schools. The Medical Division of the Eastman Kodak Company keeps in close touch with the Department and has just published an article on the Department's photographic techniques, illustrated by 27 pictures taken by Chester F. Reather.

An especially interesting example of the way in which fundamental scientific research is applied to practical ends is presented by the range-grass experiments that have been in progress under the leadership of Jens Clausen at the Department of Plant Biology during the past decade.

For many years, before this project was begun, the Department had carried on systematic studies of the way in which new plant species evolve. It had learned that the tempo of plant evolution varies greatly. At one extreme it observed the slow accumulation of hereditary differences through mutation, gene exchange through hybridization, and natural selection, which ultimately may result in the development of new species. At the other extreme it observed the sudden creation of a new species through the addition of all the chromosomes of distinct species following hybridization—a process called amphiploidy. The production of three amphiploid Madiinae in the Department's laboratory prompted it to inquire into the requirements for a successful amphiploid combination. With the three thoroughly analyzed cases of the Madiinae as a background, it made an analysis of many other well documented cases reported in the literature on the subject, in order to determine what principles govern the production of amphiploids and the criteria for their success when they appear in nature.

The investigation answered important questions regarding the evolutionary pat-

tern of many genera of plants. But its practical significance lay in the economic value that must ultimately flow from the controlled production of amphiploids. Plant breeding, which aims to improve crops by creating new forms adapted for given conditions, is most successful if it follows the proved basic laws that govern natural evolution. Ordinary methods of plant breeding are slow, but the process can be appreciably hastened where amphiploidy is feasible. Amphiploids have, moreover, both the vigor of the hybrid and the stability of independent species. And by combining the inheritances of forms from extreme climates and habitats it is possible to produce new species well adapted for intermediate environments.

A program for applying the basic knowledge that had been gained through years of pure scientific research was begun by the Department in 1943 in co-operation with the Soil Conservation Service of the United States Department of Agriculture. Its immediate object was to intercross contrasting asexually reproducing species of bluegrass from contrasting climates to produce stable hybrids having new qualities especially suitable for range conditions. The Soil Conservation Service made available mature plants of their more promising nonhybrid strains for the initial selections and crossings. It also grew cultures of the resultant hybrids in direct comparison with many different grasses in the contrasting environments of its nurseries at Pullman and Bellingham in Washington, and at Pleasanton and San Fernando in California. When a superior strain has been produced and its superiority has been proved, the Service has facilities for initial seed increase, registration, publicity, and distribution to ranchers and seed companies for the production of certified seed.

The magnitude of the range-grass ex-

periments is partially indicated by the fact that more than 150,000 individual plants have been studied. Two batches of approximately 700 first-generation hybrids were obtained by selection from approximately 88,000 nonhybrid plants. The 30 constant hybrid lines now suitable for plot testing were picked from about 50,000 hybrid individuals, representing several generations, which were thoroughly studied for several years in a series of contrasting environments.

The co-operative relations on the grass program have worked out excellently; for they have made it possible for the staff members of the Institution to devote their attention to the fundamental aspects of the research, while the application has moved forward elsewhere.

There have been many other instances in which fundamental Institution research has had its practical application, and there will undoubtedly be many more in the future.

RESEARCH ACTIVITIES

The full scope of the work in the Institution's departments of research during the past year is surveyed as usual in the reports of the several directors. Here I shall present a brief review of certain undertakings of particular interest.

Occasionally a completely new approach to the problems of one field of science becomes available as a result of the development of new techniques in some other field. This has recently occurred in astronomy through the development for radar purposes of new, highly sensitive shortwave radio receivers. Older astronomical methods were limited to studies of the radiations from astronomical bodies in the visual or near-visual range of wave lengths from 0.0003 to 0.0010 millimeter. The new radio receivers permit investigations in the wave-length range from a few millimeters to several meters. Thus the radio physicists join those who have long surveyed the heavens by optical methods.

Radiations in this new radio range were first discovered coming from the sun and the Milky Way; and, as techniques have been refined, they have been observed coming from a large number of relatively small sources. With these small sources in particular, it has been very important to identify the radio source with a known astronomical object, in order to correlate the information gained from the radio observations with that derived from observations by ordinary optical methods, and so to increase our understanding of the behavior and properties of the object. Unfortunately this identification is often difficult and uncertain, since the small ratio of aperture to wave length in the radio receivers, as compared with that in an optical telescope, makes an exact determination of the position of the radio source impracticable. But the recent development of interferometer methods, by which the angular diameter of the radio source can be measured, adds another criterion for testing these identifications.

The radio observations require completely different equipment, techniques, and skills from those used for optical observations and available at Mount Wilson and Palomar. Nevertheless collaboration between the two groups of investigators is essential, in order that the new observations may be guided by the experience of those who have long studied the puzzles of astronomy. Recently, Walter Baade and Rudolph L. Minkowski, of the staff of the Observatories, have co-operated closely with groups of radio observers in Australia

and England in an effort to correlate radio observations with optical observations made at Mount Wilson and Palomar.

During the present year this co-operation has led to the quite certain identification of the strongest of these radio sources, Cassiopeia A, with a very remarkable galactic emission nebula about 5 minutes of arc in diameter. It has also led to the identification of the second strongest source, Cygnus A, with an extragalactic nebula whose structural pattern Baade interprets as probably representing two nebulae in collision. If this is the case, the colliding masses of gas should be highly excited by the impact. This hypothesis was supported by a spectrogram, obtained by Minkowski, which shows the lines of highly ionized neon (Ne v) as a prominent feature. It is no mean achievement that man, from his humble position in the cosmos, should be able to observe at all these counterparts of our own galactic system located millions of light years away in space. But to detect, locate, and identify the collision of two such great stellar masses is truly a notable event in man's long experience of looking at the stars and wondering at the extent and arrangement of the cosmos. A mass of peculiar nebulous filaments has also been found corresponding in position and extent to a third radio source, Puppis A. Spectrograms obtained with the nebular grating spectrograph at the 100-inch telescope show that the random velocities of the filaments are of the order of 50 kilometers per second, but no systematic velocities of expansion are apparent.

A notable contribution was also made to the oldest of the fields of astronomy, that of the planetary system, when on September 28, 1951, Seth B. Nicholson discovered a twelfth satellite of Jupiter. Nicholson had discovered the ninth satellite of Jupiter at Lick Observatory in 1914, and the tenth and eleventh at Mount Wilson Observatory in 1938. The present satellite was found on a series of plates which were being taken to obtain new positions of the previously discovered satellites in order to bring their orbits up to date. The new object is a member of the group composed of satellites VIII, IX, XI, and XII, all of which move about Jupiter in a retrograde direction with a period of about 700 days. Its diameter probably does not exceed 14 miles.

Nearly fifty years ago, before the establishment of the Geophysical Laboratory, a distinguished geologist, C. R. Van Hise, submitted a report to the Trustees of the Institution on the advisability of creating a department wherein the basic processes of geology could be subjected to experimental test. It was becoming evident that for further progress in the science of geology an essential requirement was the measurement of the properties and behavior of rock-forming minerals under conditions of temperature and pressure existing in the depths of the earth as well as at the surface. Van Hise urged in particular that the role of water in the formation of minerals and rocks should not be neglected, because many minerals are crystallized from water solutions and also because water is a normal constituent of the magmas from which igneous rocks are produced.

In the years that have passed since the establishment of the Geophysical Laboratory and the initiation of work there in the field of experimental petrology, a great store of valuable information has been accumulated on the conditions under which rocks melt or under which magmas and other solutions crystallize; but by far the greater part of this information has referred only to mixtures not containing water. The experimental difficulties en-

countered in combining high temperatures with the high pressures required for retaining water or volatile ingredients in the mixtures to be studied are formidable; but happily we can now report that progress is being made at an unexpectedly rapid rate in the line of experimentation that was given primary emphasis at the beginning of the Laboratory program. New apparatus and methods have permitted measurements to be made quickly and easily at pressures up to 4000 atmospheres concomitant with temperatures up to 800° C. One of the results of the application of the new techniques is that the great and difficult problem as to how granite can be produced from a magma can now be considered solved. As a result of the methods and considerations mentioned in last year's report, we now know rather precisely the temperature at which granite rocks solidified; and we are able to comprehend the remarkably narrow range in mineral composition of the granites of the earth's crust.

An important factor in the formation of mineral deposits is the solvent power of steam when superheated above its critical point so that it cannot condense to liquid, and subjected to pressures of many thousands of pounds. Thus highly compressed until its density is comparable to that of liquid, steam dissolves mineral constituents in significant quantity. Consequently the gas which must be considered is not water alone, but water maintaining in gaseous solution other substances which profoundly influence subsequent processes of magmatic differentiation.

The measured interval during which the earth rotates once on its axis provides our unit of time. But astronomical observations have shown that the earth as a clock may accumulate an error of some seconds,

either fast or slow, in a period of only ten or twenty years.

In the late 1920's, Benjamin Boss, former Director of the Department of Meridian Astrometry of the Institution, while cataloguing positions and motions of about 30,000 stars at Dudley Observatory, concluded that there were yearly, seasonal, and perhaps even daily changes in the rate of the earth's rotation. He found that his results correlated with the hourly and monthly frequencies of earthquakes and seasonal changes in the height of tides. Although there have been numerous similar studies by many writers dealing with changes in the moment of inertia and angular momentum of parts of the earth, only a fraction of the year-to-year changes in rate of rotation could be accounted for by atmospheric and crustal changes.

Now, as a result of investigations carried on by E. H. Vestine of the Department of Terrestrial Magnetism, it appears likely that a major cause of the year-to-year changes in the rate of the earth's rotation has been found. The experimental evidence shows that the yearly rate of westward drift of planetary features of the geomagnetic field since 1900 was larger when the earth turned faster and smaller when the earth turned slower. These features of the geomagnetic field are considered to be caused by the earth's molten liquid core. The changes in the westward motion of the core, though small and difficult to detect since the accumulated amount in 30 years is only a few degrees, can provide substantial changes in the angular momentum of the core, which must be compensated for by motion of the crust and mantle. This provides an unforced explanation of the major changes in the rate of the earth's rotation derived by Brouwer for the past 50 years.

A project being jointly carried on by the

Geophysical Laboratory and the Department of Terrestrial Magnetism involves new procedures for measuring the ages of igneous intrusive rocks, especially rocks that were formed long before the first fossils were laid down in the Cambrian epoch. The feeble radioactivity of uranium and thorium, giving rise to isotopes of lead, enlarged the geological time scale early in this century to our present estimate of several billion years for the geological age of the earth. This early work was based on the highly specialized deposits represented by uranium and thorium ore bodies. One may feel uncertain about the freedom from leaching of such specialized deposits over a sweep of time as long as two billion years, but there is no such uncertainty with regard to a buttress of granite a hundred miles long.

During this year methods have been worked out for measuring the ages of samples of virgin granite and of unaltered rocks found in close association with such massive igneous monoliths as, for example, the Sierra Nevada range. These methods involve the isolation of various separate kinds of mineral, each containing its appropriate microscopic amounts of thorium, uranium, lead, rubidium, strontium, and potassium, as it was laid down in the distant past. Chemical and physical methods utilizing ion-exchange resins, radioactive counters, and the mass spectrograph, and new procedures such as isotope dilution for quantitative measurements at microgram levels, have enabled the staff members of our two laboratories to measure the ages of several igneous deposits utilizing only these micro constituents. These new methods contrast with the older methods of seeking ore bodies for age measurements, and the measurements made by them are relatively free from the possible effects of changes in the chemical constituents of rock by solution and other processes long after the original igneous activity.

Within the next few years these new methods should permit cross checks of the ages of particular igneous deposits by independent measurements of different radioactive chains in the same rock masses. Thus far there has been almost no opportunity for independent checks on the uranium-lead ages hitherto measured. The importance of such checks is perhaps illustrated by the fact that the first series of measurements on pure minerals using the rubidium-strontium procedures at microgram levels and avoiding the use of optical spectrochemistry has given age figures half again as large as those found in current reference works. The oldest known rocks are in Canada and South Africa, and the new measurements indicate ages well in excess of three billion years.

The Department of Plant Biology during the past year has taken an active part in studying some of the problems of the alga Chlorella, which was used in the Chlorella pilot plant set up in 1951 and operated by Arthur D. Little, Inc., Cambridge, Massachusetts. Many simple fundamental questions about the growth requirements of Chlorella had to be answered for the intelligent design and operation of a large-scale culture plant. Such things as the particular strains of algae used, the temperature, the chemical composition and the turbulence of the solution in which they are suspended, the supply of carbon dioxide, and the brightness of the light which causes them to grow-all have strong, interrelated, and only partially understood influences upon the growth rate.

In our study of *Chlorella*, we have been fortunate in having the co-operation of Jack E. Myers of the University of Texas and Robert W. Krauss of the University of

Maryland, two specialists in the field of algal physiology, whose experimental work has contributed to the transformation of large-scale algal culture from an art for those with green thumbs to a field in which scientific information can be applied to give reasonably predictable results.

Some experiments intended to help the development of practical algal culture have also been of more general interest because they bear upon the nature of the process of photosynthesis, which is common to all green plants. In particular, the rate of photosynthesis in Chlorella has been studied by Bessel Kok with intermittent light over ranges of intensities and of flashing schedules that were previously unexplored. It has become clear that the time required for the nonphotochemical step of photosynthesis to run its course is very different after short flashes from what it is after long flashes of light have been given the cells. The optimum temperature for Chlorella growing in bright sunlight has been found by Harold W. Milner to be higher than that previously found in indoor experiments, provided the culture is kept cool at night. Constantine Sorokin, working under Myers at Texas, has isolated a strain of Chlorella that will tolerate a much higher temperature than the common variety, and fortunately it can also use brighter light. Its growth rate in thin cultures at its optimum temperature of 39° C is twice that of the strain used in the pilot plant. Its behavior in the higher culture densities required in practice is being investigated.

The production of pound lots of dried algae of eight strains of *Scenedesmus* by Krauss at the University of Maryland is making possible the evaluation of these algae as possible sources of rare chemicals. Krauss is also testing, on a smaller scale,

numerous different kinds of algae for their potential use in mass culture.

Edwin A. Davis has found urea to be an excellent source of nitrogen for the growth of algae. He has also clarified the relations existing between cell division and the growth in size of individual cells. These two processes are very differently influenced by temperature and illumination, and it is possible to obtain cell division at night, during the day, or both day and night by the control of temperature.

It was anticipated from photosynthesis experiments of long standing that the provision of adequate turbulence in a culture exposed to bright light could, by rapidly shifting the cells from light to darkness, make possible a more efficient utilization of high light intensities. In a cylindrical chamber with a spinning rotor Davis found a 71 per cent increase of yield attributable to high turbulence.

During the past year Jens Clausen, William M. Hiesey, Paul Grun, and Malcolm A. Nobs produced about 400 new hybrids between different species of bluegrass. Some of these combine the hereditary characteristics of four distinct species. The purpose of the new crossings is partly to explore the evolutionary structure of the highly complex genus Poa, to which the bluegrasses belong, and partly to determine further possibilities for synthesizing new forms that may be superior as range grasses. Approximately 30 hybrid strains have been selected from the large number under study during the past eight years and are now ready for testing by the United States Soil Conservation Service in plots approximating agricultural and range conditions.

A basic study of the genetic and ecologic systems that characterize natural races and species is being steadily advanced, using the milfoils or yarrows as an example. Much of the great diversity found in natural populations can be matched by segregating second-generation progeny of hybrids between a few key races originally from contrasting climates. Knowledge of chromosome numbers in this complex group of plants has been greatly extended by Friedrich Ehrendorfer, of Vienna, who has studied forms from both North America and central Europe.

Recognizing the need for basic information on the comparative physiology of plants that are successful in contrasting kinds of climate, as an aid to the understanding of natural selection and evolution, Hiesey and Milner are co-operating in a program designed to explore this little-known field. Current work is being directed toward the study of the effects of temperature on rates of respiration and photosynthesis in different ecologic races.

Recent studies by Ralph W. Chaney of vegetation in many parts of the northern hemisphere indicate that forests of today have a pattern of distribution similar to that of the Eocene forests some sixty million years ago, but that they now occupy areas some twenty degrees of latitude farther south. Their shifts in position seem to have resulted from major climatic trends rather than from wandering continents and poles. Any interpretations of modern vegetation in relation to existing climate must include full consideration of the composition and distribution of the ancient forests preserved in the fossil record.

At the Department of Genetics, studies conducted by M. Demerec on the induction of mutagens in the bacterium *Escherichia coli* have progressed to a stage where he is able to postulate a sequence of events occurring in cells treated with a mutagen.

All the evidence indicates that the mutagens do not produce a direct effect on genes, but that their action is indirect, probably through some change in the cell metabolism, which in turn affects the genome. Different members of the genome react differently to conditions in cells created by the treatment. Some may be completely unaffected (mutagen-stable), whereas others may be affected to various degrees. The indications are that the conditions produced by a mutagen may persist in treated cells until they divide, and in the meantime may be modified by the application of certain other agents.

Investigations by Evelyn M. Witkin of some effects of temperature on mutability in *Escherichia coli* have revealed that the rate of spontaneous mutation responds to temperature changes according to the Van't Hoff rule, and that an intimate relation exists between spontaneous mutation and the process of cell division. They have also shown that the yield of mutations induced by ultraviolet light can be drastically affected by temperature during a critical period following irradiation.

In collaboration with M. Delbrück of California Institute of Technology, N. Visconti has formulated a quantitative theory of genetic recombination in bacteriophage. This theory postulates the transformation of phage into a vegetative form upon infection of the bacterium.

Alfred D. Hershey has established that a typical bacterial virus consists of a membrane (chiefly protein) containing almost pure nucleic acid. The function of the viral membrane in infection of the bacterial cell begins with the anchoring of the viral particle to the cell wall and quickly ends with the injection of the viral nucleic acid into the cell. Multiplication of the virus is probably a much simpler biochemical process than has been supposed; in the

initial stages it may be exclusively the synthesis of a specific kind of nucleic acid.

Investigation by Berwind P. Kaufmann has demonstrated that the shrinkage of chromosomes produced by treatment of cells with pepsin is due to the contraction of a partially hydrolized nucleoprotein gel, and not to selective action of the enzyme on a matrix protein as was previously believed.

An enzyme capable of breaking down isolated but not intact intracellular desoxyribonucleic acid has been discovered in onion root tips by Margaret R. McDonald. She has also found that one possible mechanism for the inactivation of dilute solutions of trypsin by X rays is the interaction of the trypsin molecules with hydroxyl radicals resulting from ionization of water. Hydrogen peroxide does not appear to be involved in the inactivation process.

Barbara McClintock has reached the conclusion from her studies on maize that at least two classes of functional genetic unit are carried by the chromosomes: one of them, the gene, potentially capable of determining a particular course of cellular reactions: the other associated with the control of this potential action. Unlike the gene, which occupies a fixed position in the chromosome, the controlling units may move from one location to another. When such a unit is inserted adjacent to a known gene, it exercises its specific mode of control of the time and degree of genic action, regardless of the type of potential action of the gene itself. McClintock has been able to study the operation of the same controlling unit at a number of different gene loci, and, conversely, of different controlling units at the same gene locus.

In terminating a long series of studies on induced resistance to transplanted leukemias in mice, E. Carleton MacDowell reports the separation from leukemic cells of an X factor, whose action upon the host is so rapid that a small dose of leukemic cells inoculated at the same time may eventually be suppressed. But the inoculation of the extracted X factor only initiates the host's reaction, which is speeded up by increasing amounts of the X factor provided by the inactivation of some of the leukemic cells and is still further augmented by the inactivation of the descendants of other leukemic cells. This process is interpreted as an unexpected rapid and kinetic form of active resistance. The process is believed to underlie the other procedures by which such resistance has been induced and to operate whenever a challenging dose of leukemic cells is given, even after the host has developed enough resistance to survive massive doses.

At the Department of Embryology the activities of the year included two important pieces of morphological study. George W. Bartelmez reports the discovery that there is extensive formation of neuralcrest cells by the early embryonic eye (optic vesicle), paralleling the well known formation of such cells by the primitive lower brain and spinal cord. This observation throws light on the origin of the pigmented cells which appear early in the development of the eye. Theodore W. Torrey, a guest investigator, has thoroughly reinvestigated the development of the earliest human kidney rudiments. His findings not only put a great deal of formerly incomplete and partly conflicting information into orderly sequence, but also show that certain supposed peculiarities of the early excretory organ (pronephros) in man do not exist. In this respect, as in many other details of embryonic development, our species is more like other mammals than has sometimes been thought.

S. R. M. Reynolds, who returned from

a year's visit to Oxford, brought back with him a highly instructive collection of X-ray motion pictures illustrating the fetal circulation in the sheep, obtained with the collaboration of the Nuffield Institute for Medical Research. These pictures clearly show the path of blood flow before and after closure of the ductus arteriosus, the channel through which in fetal life the unexpanded lungs are by-passed. A striking increase is seen in the rate of blood flow through the lungs when respiration begins. A remarkable deduction from the X-ray motion pictures of the umbilical arteries is that arteries in general do not expand and contract laterally to any measurable degree under the influence of the pulse.

Arpad Csapo and the Director, George W. Corner, continued with success their investigation of the physiology and biochemistry of uterine muscle. Their most important result during the year was the demonstration that the "staircase phenomenon," long known in heart muscle, occurs also in uterine muscle, and furthermore that the slope of the staircase is reversed by treatment with the corpus luteum hormone, progesterone. This discovery, which points to an essential role of potassium ions in uterine muscle contraction, hints that one of the properties of progesterone is the ability to modify the movement of potassium in and out of the muscle cell. The investigation is thus getting very close to the fundamental problems of the specific manner in which hormones exert their extraordinary physiological effects upon their target organs.

At the request of the United States Army Medical Corps, the Department of Embryology has continued its study of the permeability of capillary blood vessels with respect to two of the most promising blood substitutes now under consideration for military and civilian use in combating shock from loss of blood.

During the past year the Department of Archaeology completed its first field season of full-scale operations under a new program of studies. It is not uncommon in the initial stages of research to find that more problems are raised than solved. This proved to be the case in the exploratory work carried on at the extensive ruins of Mayapan in northern Yucatan, which is the scene of the present activities of the Department.

Examination of the ruins, as yet unchecked by excavations of any magnitude, has brought the surprising disclosure of the apparent absence, or near absence, of archaeological remains of a period in history strongly associated with the ancient city through documentary and historical records. This disclosure has led to the review of evidence supporting certain widely held beliefs concerning the late preconquest history of Yucatan. It has commonly been thought that toward the end of the tenth century Yucatan was invaded by a band of people who brought with them a foreign culture that spread widely, and deeply affected the native Maya civilization. Their seat of authority was Chichen Itza. The ascendancy of these people supposedly lasted about two hundred years, after which time their foreign ways and ideas were gradually absorbed by the older, native culture, a process that presumably continued until the Spanish conquest.

Although the architecture and sculpture at Chichen Itza leave little doubt of the presence of the foreigners at that city, virtually no traces of that architecture and sculpture are found elsewhere in Yucatan. On the basis of strictly archaeological evidence, as contrasted with that of written

record, one is led to question the generally accepted character and duration of this period in Yucatan history. Any considerable change in this part of the historical framework involves the time and character of preceding and succeeding periods. Other evidence suggests that in later times the

native civilization exhibited more rather than fewer foreign elements, contrary to what has been supposed. The work of the past season has thus resulted in the formulation of a series of problems that are fundamental to the studies of the Department.

STAFF

Honors and recognition for contributions to the advancement of knowledge have come as usual to members of the Institution's staff, and it is with satisfaction that I bring these to your attention. Dr. Ira S. Bowen, Director of the Mount Wilson and Palomar Observatories, was invited to give the Halley Lecture at Oxford University, England, in May 1952. November 8, 1951, the Mineralogical Society of America presented an award to Dr. Orville F. Tuttle, petrologist of the Geophysical Laboratory, for his work on the variable inversion of quartz. Dr. Merle A. Tuve, Director of the Department of Terrestrial Magnetism, received, in recognition of his activities in fields adjacent to science, the degree of Doctor of Laws honoris causa from Augustana College (Sioux Falls), of which Dr. Tuve's father was president for many years. From Joint Task Force Three, Los Alamos, New Mexico, Dr. George R. Wait, retired staff member of the Department of Terrestrial Magnetism, received a Certificate of Achievement for meritorious service. Dr. Herman A. Spoehr, Chairman Emeritus of the Department of Plant Biology, served as President of the Pacific Division of the American Association for the Advancement of Science. Dr. William M. Hiesey, on invitation, presented a paper at the Colloque International sur l'Evolution chez les Plantes held in Paris. On April 26, 1952, Dr. M. Demerec, Director of the Department of Genetics, was elected a mem-

ber of the American Philosophical Society; and on April 29, 1952, Dr. Berwind P. Kaufmann, staff member of the Department, was elected to membership in the National Academy of Sciences. Dr. George W. Corner, Director of the Department of Embryology, was elected Honorary Fellow of the Royal Society of Edinburgh, and in May 1952 he was invited to give the Huxley Lecture at Charing Cross Hospital Medical School in London. Mr. Richard D. Grill, photographer at the Department of Embryology, received a Certificate of Merit for his entry of three pictures of a human fetus in utero in the Sixteenth Rochester International Salon of Photography. In September 1951 Dr. A. V. Kidder, retired Director of the Department of Archaeology, received a degree from the National University of Mexico of Doctorado honoris causa. Dr. E. A. Lowe, Research Associate of the Institution with the Institute for Advanced Study, has been elected a corresponding member of the Bavarian Academy of Munich and the Accademia dei Lincei of Rome.

Two distinguished members of the Geophysical Laboratory who have worked for many years to advance fundamental knowledge on the evolution of rocks in the earth's crust have retired: Dr. Norman L. Bowen on June 30, 1952, and Dr. Leason H. Adams on July 31, 1952.

Dr. Bowen has been associated with the Laboratory since 1910 except for two breaks in service, one for a year as professor of mineralogy at Queen's University in Kingston, Ontario, and another for ten years as professor of petrology at the University of Chicago. He has been concerned from the beginning with studies in phase equilibria in mineral systems at high temperatures, and gained his reputation among petrologists with the publication of The Evolution of Igneous Rocks in 1928. This monograph is recognized as a classic in the field. Dr. Bowen has been pre-eminent in bringing chemical knowledge to bear on the problems of geology through new techniques of investigation in the laboratory and correlation of laboratory findings with field evidence. He expects to continue his research and will also prepare for publication the results of his work for the past few years on the melting relations of granite in the presence of water and its application to geological problems.

Dr. Adams has been with the Laboratory almost from its beginning. He joined the staff as a physical chemist in 1910, became Acting Director in 1936, and Director on January 1, 1938. With E. D. Williamson, he worked out and published in 1920 the theory of glass annealing that laid the foundation for modern methods of annealing. Always interested in the chemistry and physics of the processes of the earth, Adams' investigations on the effects of high pressures on rocks and rock-forming minerals and his studies of chemical systems have thrown considerable light on

the internal constitution of the earth and on the influence of pressure on the critical temperature of magnetization for iron and other materials. Dr. Adams expects to continue his writing, with headquarters in the Administration Building of the Institution.

On October 1, 1951, Dr. George R. Wait retired after thirty-one years of service as a member of the research staff of the Department of Terrestrial Magnetism. He had, with S. J. Mauchly, played an important part in the discovery of the now well known universal wave in the diurnal variation of the earth's electric field. Later, while in charge of the Institution's magnetic observatory at Watheroo, Western Australia, he initiated investigational work on earth currents and atmospheric electricity in that part of the globe. His research in atmospheric electricity included primarily study of the production and destruction of atmospheric ions, and investigations with Dr. O. H. Gish of the origin and maintenance of the earth's electric charge. The results of these investigations furnish the strongest evidence thus far obtained in support of the theory previously advanced that the thunderstorms of the globe are responsible for the return negative charge to ground. Dr. Wait is now engaged as a scientific investigator with the Geophysics Research Division, Air Force Cambridge Research Center, Cambridge, Massachusetts.

James W. Wadsworth

The Institution lost a distinguished member of its Board of Trustees on June 21, 1952, at the age of seventy-four. Elected to membership twenty years ago on December 9, 1932, James W. Wadsworth served as a valued adviser both on the Institution's Auditing Committee and on its Committee on Historical Research,

now the Committee on Archaeology. He always took an enthusiastic interest in public affairs. Succeeding Elihu Root, he served for two terms in the Senate and later nine terms in the House of Representatives. During his thirty years in Congress he incessantly devoted himself with vigor and courage to the cause of national

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security, beginning even before World War I, when he advocated universal military training. Recognition of his contribution to the development of our military security came in 1951, when he was appointed a member and later chairman of

the National Security Training Commission. The report of this Commission furnished the basis for current proposals for universal military training. Mr. Wadsworth's death will be keenly felt both by the nation and by the Institution.

FINANCES

Statements showing the financial position of the Institution at June 30, 1952, together with a certificate of Haskins and Sells, Certified Public Accountants, appear in the report of the Executive Committee.

As indicated by these statements, income from the Institution's investments for the past fiscal year amounted to \$1,888,569.58. This was about \$95,000 less than the investment income received for the preceding fiscal year.

Although income from investments fell below that of the preceding year, total appropriations and allotments approved by the Board of Trustees and the Executive Committee during the year were also about \$92,000 less than in the preceding year, with the result that actual investment income exceeded expenditures approved during the year by about \$110,500. This is the net amount of investment income which was credited to reserves during the year.

The yield at book value of securities held during the year was 4.36 per cent, and at market value on June 30, 1952, the yield was 3.68 per cent. The book value of in-

vestments at the end of the year was \$43,390,283, an increase of \$788,524 over the close of the preceding year; and the market value of the portfolio increased from \$49,273,504 at June 30, 1951, to \$52,546,226 at June 30, 1952.

Diversification of the portfolio changed but slightly during the year. At market value, 42.8 per cent was in bonds, 49.2 per cent in common stocks, 7.8 per cent in preferred stocks, and 0.2 per cent in cash at the close of the year. Of the income from investments, 63.5 per cent came from common stocks, 27.5 per cent from bonds, and 9 per cent from preferred stocks. Thus the Institution's equity holdings continue to support a major portion of its operations. Despite slightly reduced income, it has been possible to adjust expenditures with the result that a well balanced research program has been continued and a reasonable sum has been credited to reserves, as has been our policy for some years, in the light of the proportion of equities in the portfolio.

VANNEVAR BUSH

REPORTS OF DEPARTMENTAL ACTIVITIES AND CO-OPERATIVE STUDIES

ASTRONOMY

Mount Wilson and Palomar Observatories

TERRESTRIAL SCIENCES

Geophysical Laboratory

Department of Terrestrial Magnetism

BIOLOGICAL SCIENCES

Department of Plant Biology
Department of Embryology
Department of Genetics

ARCHAEOLOGY

Department of Archaeology

REPORTS OF RESEARCH ASSOCIATES



MOUNT WILSON AND PALOMAR OBSERVATORIES

OPERATED BY THE CARNEGIE INSTITUTION OF WASHINGTON AND THE CALIFORNIA INSTITUTE OF TECHNOLOGY

Pasadena, California
IRA S. BOWEN, Director

OBSERVATORY COMMITTEE

IRA S. BOWEN, Chairman ROBERT F. BACHER JESSE L. GREENSTEIN EDWIN P. HUBBLE PAUL W. MERRILL ERNEST C. WATSON

The Palomar Observatory was made possible by a grant of \$6,000,000 to the California Institute of Technology in 1928 by the International Education Board. This was supplemented by additional grants totaling \$550,000 by the Rockefeller Foundation to cover increased costs of construction after World War II. These funds had all been expended by December 1951 and fortunately were sufficient to finish the telescope and practically all its auxiliary equipment.

In one sense, however, the telescope will never be completed, at least not until it ceases to operate. The telescope proper is basically an instrument for collecting and concentrating to a focus the light from a star or nebula. Once collected, the light is measured with a photometer, analyzed with a spectrograph, or recorded with a photographic plate. Whereas the main light-collecting element, that is the 200inch mirror and its mounting and control mechanism, will normally continue to function without major modification for a long time, the auxiliary equipment for the study of the light will require frequent modification and even replacement as new advances in optics, electronics, and related fields render the present equipment obsolete. Indeed, entirely new types of instrument may be needed for kinds of measurement that are not even contemplated at the present time.

Such revisions and replacements have occurred regularly with the auxiliary equipment on Mount Wilson. Complete new coudé and Cassegrain spectrographs were constructed in the late 1930's for the 100inch telescope to utilize the newly developed schmidt cameras and thereby to obtain greatly increased speed and resolving power. Since 1945 a Newtonian spectrograph and a photometer using the new photomultiplier-tube techniques have been built for this telescope. A new Cassegrain grating spectrograph is now under construction for the 60-inch telescope. As soon as this is completed, a thorough revision of the 100-inch coudé spectrograph is planned. In addition, dozens of minor revisions and replacements of small parts of equipment take place each year.

The 60-inch and 100-inch telescopes on Mount Wilson are now 44 and 34 years old, respectively. However, by this continued renewal of the auxiliary equipment that uses the light collected it has been possible to keep these telescopes numbered among the most effective instruments for astronomical research that are available today.

It is of course expected that similar modifications and replacements will be neces-

sary for the equipment used at the Palomar Observatory. Indeed, modifications of some of the auxiliary equipment completed first had already taken place before all items of equipment had been finished.

Obviously, extensive design and optical and machine-shop facilities must be maintained for this program of modification and new construction of auxiliary equipment. The facilities of both the Palomar and Mount Wilson Observatories have been undergoing gradual reorganization to take advantage of gains in efficiency made possible by the start of the joint operation of the Observatories in 1948. Thus, with the completion of most of the major optical parts for the Palomar Observatory in 1949 the optical shop on the campus of the

California Institute was closed down and all optical work for both observatories was concentrated at the optical shop on Santa Barbara Street. Likewise, as retirements have occurred in the staff of the Mount Wilson machine shop at Santa Barbara Street, no replacements have been made, but more and more work has been sent out to the Central Shop Facility on the California Institute campus. This Facility has taken over the shop which was built for the construction of the 200-inch telescope. Fortunately the tapering off of the construction at Palomar has made available the necessary capacity to handle the Mount Wilson construction, which eventually will be very largely combined there with that of the Palomar Observatory.

OBSERVING CONDITIONS

After seven years in all but one of which the precipitation on Mount Wilson was far below normal, a very plentiful rainfall was received, the recorded fall of 56.84 inches being over 50 per cent above normal. Solar observations were made on 320 days between July 1, 1951 and June 30, 1952. During this period observations were made with the 60-inch telescope on 289 nights and with the 100-inch on 292 nights.

SOLAR RESEARCH

Solar Photography

Solar observations were made by Cragg, Hickox, Nicholson, Parker, Richardson, and Roques. The numbers of photographs of various kinds taken between July 1, 1951 and June 30, 1952 were as follows:

Direct photographs	608
Ha spectroheliograms of spot groups,	
60-foot focus	760
Ha spectroheliograms, 18-foot focus	870
K2 spectroheliograms, 7-foot focus	68,000
K2 spectroheliograms, 18-foot focus	900
K prominences, 18-foot focus	939

SOLAR ACTIVITY

The magnetic classification and study of sunspots and related phenomena have been

continued by Nicholson, Parker, and Cragg. Co-operative programs have been carried out with the United States Naval Observatory, the Observatory of Kodai-kanal, the Meudon Observatory, the Department of Terrestrial Magnetism of the Carnegie Institution, and the Central Radio Propagation Laboratory of the National Bureau of Standards.

During the calendar year 1951, solar observations were made at Mount Wilson on 329 days, one of which, August 27, was without spots. The total number of spot groups observed in 1951 was 294, which was 83 less than in 1950.

The northern hemisphere with 158 spot groups was more active than the southern with 136. Following the minimum of 1944,

sunspot activity developed more rapidly in the southern hemisphere than in the northern until a maximum was reached in 1947. Activity in the northern hemisphere continued to increase until a maximum was reached in that hemisphere in 1949. Since 1949 activity has declined in both hemispheres, with the northern hemisphere the more active each year.

The monthly means of the number of spot groups observed daily for the past two and one-half years are shown in table 1.

TABLE 1

Month	Daily number						
	1950	1951	1952				
January	8.7	4.0	2.8				
February	6.9	4.5	2.0				
March	8.7	4.2	2.0				
April	6.9	5.7	2.2				
May	8.1	7.0	2.2				
June	6.2	5.5	3.8				
July	6.2	5.4					
August	7.0	4.9					
September	4.7	6.4					
October	5.4	5.2					
November	4.5	5.2					
December	4.1	3.4					
Yearly average	6.5	5.0					

The year 1951 was marked by a decided increase in solar activity following the rapid decline in 1950. After the peak in May 1951, sunspot activity declined steadily until February and March 1952. Since then it has been increasing slightly toward a new secondary peak like that of May 1951.

Thirty-two solar flares of intensity 1, five of intensity 2, and none of intensity 3 were recorded at Mount Wilson during 1597 hours of observing. The average number of flares recorded per 100 hours of observing was 2.3 in 1951, 5.5 in 1950, and 6.8 in 1949.

SUNSPOT POLARITIES

Magnetic polarities in each spot group have, so far as possible, been observed at least once. The classification of groups observed between July 1, 1951 and June 30, 1952 is indicated in table 2. "Regular" groups in the northern hemisphere are those in which the preceding spot has S (south-seeking) polarity and the following spot N polarity; in the southern hemisphere the polarities are reversed.

TABLE 2

Hemisphere	Regular	Irregular	Unclassified
North	101 99	5 2	32 24
Whole sun	200	7	56

LARGE-SCALE MOTIONS IN THE PHOTOSPHERE

Large-scale motions in the solar photosphere have been studied by Richardson and Dr. Martin Schwarzschild by means of a statistical analysis of the latitude motions of 1800 recurrent spot groups. The analysis gave an apparently significant oscillation with a 22-year period, in the sense that for a cycle such as the one which started in 1914 the motions were directed toward the equator, whereas for cycles such as that which started in 1924 the motions were poleward. The semiamplitude of the velocity of oscillation was 0°0043 per day or 60 cm/sec, with a probable error of ocooo per day. This velocity variation in a 22-year cycle corresponds to a semiamplitude in latitude displacement of 5°5.

Motions in the Hydrogen Chromosphere

Spectroheliograms have been obtained by Münch and Richardson for a study of the hydrogen chromosphere similar to that made of the size and motions of the granulations in the photosphere. The spectroheliograms show a mottled structure as far as 1 A from the center of $H\alpha$. Preliminary results indicate that the mottlings change form with about the same rapidity as the granulations.

PLANETS AND SATELLITES

While systematically photographing the known faint satellites of Jupiter with the 100-inch reflector, Nicholson discovered a new one, the twelfth, on September 28, 1951. The new satellite, which is a member of the retrograde group, VIII, IX, XI, and XII, with periods of about 700 days, is fainter than any of the others. Its diameter is probably not greater than 14 miles.

A very limited program for the photography of planets at the coudé foci of the

large reflectors has been started. A 35-mm motion-picture camera has been developed for this program by Pettit and Richardson and still-camera equipment by Humason. A few very satisfactory photographs have already been obtained with both sets of equipment. Because of the limitations imposed by seeing conditions, however, it cannot be hoped that any substantial gains in resolving power will be made over that already achieved with smaller instruments.

STELLAR SPECTROSCOPY

During the report year more than 1800 spectrograms were taken with the 60-inch and 100-inch telescopes on Mount Wilson. More than 270 spectrograms have been obtained during this period with the 200-inch Hale telescope at Palomar Mountain in spite of the fact that this instrument has been used largely for long exposures of objects too faint to be observed at Mount Wilson.

A few of these spectrograms have been taken for the determination of radial velocities of stars and nebulae. The great majority of the plates, however, were obtained for studies of the chemical composition and the physical conditions such as temperature, pressure, magnetic field strength, and internal motion of the stellar atmospheres. For some of the stable stars quantitative results can now be obtained. On the other hand, in the more unstable stars such as the variables, and the stars with very extended atmospheres such as the shell stars, these problems are so complicated that only qualitative interpretations can in general be attempted at this time.

Long-Period Variable Stars

The spectrograms that, many years ago, led to the recognition of S-type stars as a third type of red star paralleling the M-type and N-type branches of the stellar sequence were taken with the 100-inch and 60-inch telescopes with a dispersion of about 35 A/mm. More recently a special study of R Andromedae, a typical S-type variable, was made with the relatively high dispersion of 10 A/mm. The 200-inch telescope has made it feasible to obtain observations of additional S-type stars with the higher dispersion. During the year Merrill has made a survey of spectrograms of eight stars taken by Bowen with the 200-inch telescope. These spectrograms, in addition to providing detailed descriptions of S-type spectra, revealed a remarkable and rapidly changing behavior of certain bright lines of vanadium and chromium in the spectrum of R Cygni. Spectrograms of R Cygni and x Cygni in future years promise to reveal several phenomena of great physical interest.

An unexpected result was the finding in S-type spectra of dark lines of neutral tech-

netium. No completely stable isotope of this element is known on earth, the most nearly stable having a half-life of less than a million years. Hence it is surprising to find this element in stellar atmospheres. Of several explanations, none is compelling: (1) a stable isotope exists although not yet found on earth; (2) the star somehow manufactures technetium as it goes along; (3) S-type stars represent a comparatively transient phase of stellar existence.

Sample spectrograms, with dispersion 9 A/mm, of 10 long-period variables having high velocities were taken during the year with the 200-inch telescope. These plates show that the absorption spectra do not differ markedly from the spectra of ordinary giant stars. The bright lines appear to behave in about the same way as those of other Me variables of similar types and periods.

Five spectrograms, dispersion 10 A/mm, were obtained of the long-period variable R Hydrae during the time when its light was increasing after minimum. Near minimum, ground-level lines of Ca 1, Al 1, and $K_{\rm I}$ were extraordinarily intense. Bright hydrogen lines appeared in low intensity about 50 days after minimum, with displacements algebraically greater than those measured previously at maximum light. The emitting hydrogen atoms appear to have an outward acceleration until they disappear before the next minimum. The dark lines, on the other hand, yield about the same velocity shortly after minimum as they do at maximum.

A study by Merrill of R Leonis during the declining phase was based mainly on six spectrograms taken with the 200-inch telescope, fairly well distributed over the interval from maximum light to minimum. The results increase our knowledge of the very complicated spectroscopic behavior of long-period variables as they approach minimum light. It now appears

that under favorable circumstances the 200inch telescope could follow the spectrum of R Leonis with dispersion of 9 A/mm right through minimum light. It remains important to obtain more extensive data at this phase.

Short series of spectrograms of four additional long-period variables have been obtained with the 200-inch telescope. With minor variations the behavior of the bright lines in all four stars conforms to the complex pattern previously observed in o Ceti and R Leonis.

Absorption and emission lines of the long-period variable star o Ceti were measured by A. H. Joy on more than 100 spectrograms having dispersions from 2.3 to 10 A/mm. These coudé exposures were made during the interval 1935-1951 by Merrill and Joy at the 100-inch on Mount Wilson and by Bowen and O. C. Wilson at the 200-inch Hale telescope on Palomar Mountain. The high accuracy obtained (mean error for a single line 0.5 to 1.0 km/sec) makes it possible to check the low-dispersion results published in 1926 and to examine in detail the behavior of this typical Me variable star at different phases and cycles.

Though the earlier results which indicated velocity variation in o Ceti are largely confirmed, it now seems certain that marked irregularities occur and that fluctuations in the maximum absorption-line velocities are nearly as large as the variation within any one cycle. Higher maximum velocities of recession usually accompany the brighter maxima of light. The greatest velocity of recession (subsidence) is generally attained 40 to 50 days after maximum light.

The observed velocities of the emission strata are more regular. They are always rising with respect to the absorption strata, but at speeds decreasing from maximum to minimum light.

Buscombe and Merrill have completed a photometric study of the ultraviolet absorption lines on coudé spectrograms of o Ceti. The strong resonance lines of Al 1 and K 1 weaken at maximum light to one-third their intensities at a phase 3 mag. fainter, owing to ionization, while the H and K lines of Ca II strengthen. An electron pressure of 1010 dynes/cm2 would be consistent with this behavior. The neutral lines of elements of higher ionization potential generally appear considerably stronger near maximum. Since this effect is especially marked among the subordinate lines, an increase in excitation temperature appears to occur, along with a great decrease in opacity, at the brightest phases.

Comparisons of R Leonis (M8e) with R Andromedae (Se) indicate that absorption lines of titanium, as well as those of zirconium and adjacent elements in the periodic table, are much stronger in the Stype star. The spectrum of o Ceti shows an intermediate distribution of these elements. It is suggested that the over-all abundance of titanium may be the same for M and S stars, but that this element is less readily oxidized in stellar atmospheres having excessive amounts of zirconium.

CEPHEID VARIABLE STARS

Bappu has taken spectrograms at different phases of the 3.7-day cepheid RT Aurigae with a dispersion of 10 A/mm for the purpose of a quantitative analysis of a cepheid atmosphere having such a period. Some plates with a dispersion of 4.9 A/mm were taken at maximum and minimum phases as well as at a point halfway between these phases.

An analysis of the population II cepheid W Virginis was made by Abt using coudé plates of 10 A/mm, some of which had been taken earlier by Sanford. Light-curves in one color by Gordon and Kron and in

two colors by Whitford and Code were used for phase determination.

A new expansion is first indicated by the appearance of H emission lines presumably formed deep in the atmosphere. Later the outward-moving material forms a set of absorption lines similar to those of an Ftype star. At its first appearance the new set of absorption lines indicates an excitation temperature of 5400°, a relatively low turbulent velocity of 5 km/sec, and a large outward velocity of 45 km/sec. As expansion continues, the absorption lines gain in strength, and the temperature and electron pressure fall. The curve-of-growth analysis shows minimum electron pressure at maximum expansion and minimum excitation temperature shortly after it. During the ensuing contraction the electron pressure, temperature, and opacity increase rapidly. Just after maximum expansion the appearance of a new set of H emission lines indicates the start of a new outward-moving wave. There is then a period of several days during which two sets of absorption lines are present, one showing expansion, the other contraction. When two sets of absorption lines are present, the excitation temperatures are 800° to 1000° C lower for the old, contracting shell than for the new, expanding one. Finally the old set of lines fades out and disappears, completing a sequence of spectroscopic phenomena extending over about 1.45 photometric periods, in marked contrast with the behavior of population I cepheids. The turbulent velocity increases throughout the cycle until the lines finally disappear, when a maximum value of 12 km/sec is reached.

SHELL STARS

Bright hydrogen lines were frequently observed in the spectrum of Pleione from 1888 to 1903, but they were absent in 1905, not reappearing until 1938. The shell episode began in 1939 and lasted until 1951;

during this interval, numerous narrow absorption lines indicated that the star's atmosphere extended to an unusual height above the photosphere. This upper absorption level, called a shell, was probably formed by atoms which had moved up from the photosphere. The complicated atmospheric motions were studied by Merrill by measuring the displacements of lines of various elements. At first the motions were so slow that the observed radial velocity of the shell was the same as that of the star as a whole. Gradually the acceleration increased, and the outward velocity of the upper levels became appreciable. Eventually the supply of atoms from the photosphere failed, and the shell blew away. The episode illustrates several characteristics of abnormally extensive atmospheres surrounding early-type stars.

Additional spectra of Pleione have been obtained by Bappu with a dispersion of 10 A/mm. Tracings of these and of earlier plates obtained by W. S. Adams at 2.9 A/mm are being made preparatory to an analysis of the physical phenomena occurring during the evolution of the shell.

During the year, brief descriptions of the spectra of 15 shell stars have been prepared for publication by Merrill. Several of the stars have been followed for a number of years, especial attention being given to the Balmer series of hydrogen lines. In most of the stars the velocities change slowly with time. The change is usually greater for the metallic lines and the ultraviolet hydrogen lines than for $H\beta$ and $H\gamma$. Of the apparently stable shells, that of HD 193182 exhibits one of the most striking spectra thus far photographed. Lines in the Balmer series of hydrogen were measured down to H_{42} . Lines of ionized metals are numerous and extraordinarily sharp. Three shell spectra showing narrow helium lines were studied; these shells are probably similar to the commoner type showing numerous metallic lines, but differ in having higher excitation.

Studies of numerous spectrograms of the shell stars 48 Librae and HD 33232 are under way.

Greenstein has made studies of the F5 giant star π Pegasi, which is probably a member of the Ursa Major Cluster. It proved to have a shell spectrum of an unusual type. The rate of rotation is large, and there are relatively sharp deep absorption cores in H and K and possibly at $H\alpha$. No emission or other shell features are detected. Thus π Pegasi is probably the coolest known shell spectrum.

A special study by Merrill of the helium line $\lambda 3888$ in the spectrum of the peculiar Be star BD +11°4673 showed the presence of from two to eight narrow absorption components having displacements of from -56 to -428 km/sec. The components vary rapidly and apparently irregularly in position and intensity. They probably arise in a layer far above the photosphere. The discrete outward motions, possibly bearing some analogy to the rapid motions of eruptive prominences on the sun, offer a promising subject for theoretical discussion.

Two faint stars, MWC 490 and HD 59771, studied because of the discovery of bright $H\alpha$ lines in their spectra, were found to be spectroscopic binaries with large ranges in radial velocity. In the spectra of both stars, displaced dark lines indicate a cloud of hydrogen expanding rapidly above a quiescent F-type reversing layer.

A faint absorption spectrum of type gM2 is measurable in the red region of the spectrum of the peculiar Be shell star HD 45910. The varying displacements of the M-type lines measured by Merrill correspond to motion in a circular orbit of period 232 days, semiamplitude 52 km/sec. The system has a curious similarity to that of T Coronae as observed by R. F. Sanford.

LATE-TYPE STARS WITH EMISSION LINES OF CALCIUM

A study of several stars with emission H and K lines has been made by Greenstein at high dispersion. The emission-line widths increase with luminosity. The superposed absorption cores are normally very sharp and near zero velocity in dwarf stars; they are broader and subject to velocity variation in the giants. The sharpness of absorption cores in the dwarfs cannot be explained by an ordinary chromospheric model for the emission. The kinetic temperature indicated by the emission lines is very high, and it is difficult to understand why cooler material capable of absorption should exist at the outer boundary of the star's chromosphere.

O. C. Wilson and Bappu are carrying out an investigation of the occurrence of emission of the calcium H and K lines in F, G, K, and M stars. The first series of plates was taken by Wilson in 1938 and 1939. The second-epoch plates of some 20 stars representative of different luminosities were taken by Bappu this year in order to detect variation of the emission features with time; several additional stars have been included in the recent group of spectrograms to extend the investigation to a larger range of absolute magnitude. Measurements are in progress of the displacements of central reversals and of the profiles and intensities of the emission features.

Abundance of Chemical Elements and Physical Conditions in Stellar Atmospheres

Greenstein has carried out extensive spectrophotometric investigations of chemical and physical conditions in the atmospheres of many stars of both normal and abnormal types. Spectrophotometry of the G stars at 2.9 A/mm has been continued and expanded. Eight stars (average type

G5) have been measured, and an analysis has been begun. Three early G stars have been added. A separate spectrophotometric study of abnormal G stars has been started: ζ Capricorni (rich in rare earths), HD 18474 (weak in CH), R Coronae Borealis (a carbon supergiant). The normal G-star program involves the measurement of 20,000 equivalent widths, and includes stars of high and low velocity.

Subdwarfs have proved to be very interesting. An outstanding feature is the weakness of all lines and of the CN absorption in the F and G subdwarfs. The very blue tenth-magnitude O star BD +28°4211 (discovered by MacRae, Fleischer, and Weston) has an extraordinary spectrum in which all lines are shallow and weak except \(\lambda\)4686. Its most striking feature, however, is its luminosity. Three good plates of 10 A/mm reveal only the weakest indication of an interstellar K line. Therefore the visual absolute magnitude must be fainter than +3 (possibly +4). The star is, then, halfway between the main sequence and the white dwarfs, with a radius smaller than that of Jupiter. It may be related to the very blue faint stars found in globular clusters by Sandage and Baade; such stars may be collapsing on the evolutionary path to the novae or white dwarfs.

Stars of very high velocity are being observed on the 4.5 and 10 A/mm dispersion. Though many of the high-velocity stars have weak *CN*, a group with strong *CN* exists. Several such G and K giants with abnormal spectra are being investigated by Greenstein.

About 150 lines suitable for spectrophotometry have been selected in the spectrum of the magnetic variable star HD 124248, and measures of equivalent width and central intensity are now being made by Deutsch on a series of coudé spectrograms taken by H. W. Babcock and himself. To the extent that the conventional curve-of-growth analysis can be applied to such an object, an attempt is being made to find relative abundance, ionization temperature, electron pressure, and turbulence as functions of phase in the cycle of variation. Concurrently, a similar analysis is being made of coudé spectra of γ Equulei, which has a magnetic field but shows no spectrum variation.

Coudé spectra of 9 Wolf-Rayet stars of both the carbon and nitrogen sequences were obtained by Bappu with the moderately high dispersion of 10 A/mm in the blue and 20 A/mm in the red. These are being examined for the detection of structure in the emission bands and also for a detailed spectrophotometric study, the main goal being the determination of the physical characteristics of the Wolf-Rayet atmosphere. Spectra of the eclipsing variable V444 Cygni were also obtained both in and out of primary minimum, in order to study the electron scattering effects in the Wolf-Rayet atmosphere. With the 60inch Cassegrain spectrographs, a low-dispersion survey of Wolf-Rayet stars brighter than magnitude 10.5, in the northern hemisphere, has been started for the purpose of spectral classification of these objects on a quantitative basis. Also spectra at different phases of the binaries HD 214419, HD 193576, HD 193928, HD 211853, HD 186943 have been taken in order to study the variation with phase of the emission-line intensity and the structure of the emission-line profiles. It is hoped that such a study will help in gaining some knowledge concerning the kinematical and physical features of the Wolf-Rayet atmosphere.

Buscombe has continued a detailed study of the spectra of the cool carbon stars, extending the measures of intensities of individual absorption lines in the red band system of the *CN* molecule to cooler stars in the N-type sequence, in an effort to make

quantitative determinations of the temperature differences as well as of the increase in abundance of *CN* in the atmospheres of these stars.

Peculiar A Stars

The visual companion of α Piscium, which is a "strontium star," has been found by Deutsch to be a spectroscopic binary exhibiting two rich, sharp-line spectra. The brighter star itself has wide lines, and there is some indication that it, too, may be a binary. Spectrum variation in the "strontium star" 21 Comae has also been studied on coudé spectra. Most of the lines in this spectrum are relatively wide, but a few unidentified sharp lines appear on some plates. Low-dispersion plates have been made at the 60-inch in an attempt to find the period of this star and of certain other spectrum variables, but no definite period has yet been found. The stars HD 107612, ω Ophiuchi, and 45 Leonis have been found to be spectrum variables with periods of the order of 2 days. HD 124224 was found to show variable lines of He I and Si II with a period of 0.52 day, the shortest period known among the spectrum variables. The contour of Mg II 4481 in this star is compatible with the hypothesis that the star rotates about an axis nearly perpendicular to the line of sight, with a period equal to that of spectrum variation.

STELLAR MAGNETIC FIELDS

Numerous measures of the periodically varying Zeeman effect and the velocity of various elements in the bright spectrum variable α^2 Canum Venaticorum by Miss Burd and H. W. Babcock have been completed, and a curve of magnetic variation has been derived.

A dozen plates of HD 133029 taken by Babcock in the interval March 31 to April 5, 1952 have been measured by Miss Burd. The star has a magnetic field which fluctuates from day to day, but without apparent regularity, sometimes reaching a polar intensity of 10,500 gauss. The polarity is always positive, and there are no obvious variations in line intensity.

Numerous other spectrograms made by Babcock with the analyzer have been accumulated for some 30 other magnetically active stars whose variations are being followed. With the coudé spectrograph of the Hale telescope, a beginning has been made on the study of magnetic stars in the northern part of the sky. Evidence of a magnetic field has been found in the 20-day spectrum variable 73 Draconis, among others.

Measurements of the stellar Zeeman effect depend directly on the analysis of elliptical polarization in the profiles of absorption lines. The initiation of such observations with the five-mirror coudé system of the Hale reflector required a new examination of the problem of compensating phase shifts due to the three oblique reflections. The theory of the phase compensator having two crossed mica plates was worked out, and a new compensator was built and calibrated in the telescope by means of a circularly polarized source.

For 1500 spectrum lines of interest in magnetic stars, Miss Burd has computed and tabulated "Z values." The Z value is the half-width of the blended Zeeman pattern as observed in a longitudinal magnetic field. This table should facilitate reduction of measures.

INTERSTELLAR LINES

Observations of the interstellar H and K lines in the spectra of distant early-type stars have been undertaken by Münch. This program is an extension of the survey work of W. S. Adams, on the same subject, to selected regions of the sky and to stars at great distances from the sun. It is planned to study the velocities, intensities,

and structure of the interstellar lines in distant stars, with a view to finding out if the distribution of interstellar gas in the galactic system presents any large-scale structural features, such as Baade has observed in M 31.

Up to date, 85 plates have been obtained. Most of them are for different stars and on a dispersion of 10 A/mm. The following stars show such unusual interstellar features that they deserve special reference:

HD 93521 ($m_v=6.89$) is a broad-line main-sequence Oo star with height z above the galactic plane of z=1300 psc. A plate taken with the 32-inch camera suggested that interstellar K might be double. Another plate with the 73-inch camera confirmed the duplicity and gave indication of a third component. Observation of the star with the 114-inch camera showed that K clearly has four components, one of which is broad enough to suggest the existence of a fifth one. This star shows, thus, that the case of ρ Leonis (z=650 psc), first studied by Beals, which Adams found to have K with three components, is not unique, and that the interstellar gas may extend to great heights above the galactic plane in special regions. A search for multiple lines in other stars in the general neighborhood of p Leonis and HD 93521 was unsuccessful.

HD 50064 (mv=8.9, E=0.40) is a highluminosity B5 star at a distance of about 3 Kpc. Although it is in a region where no stars with very strong interstellar lines were known, it shows the strongest complex K line of any thus far observed: its equivalent width is estimated to exceed 1 A, and most of this is probably of interstellar origin. This star is tentatively placed, in the diagram of the H II regions recently given by W. W. Morgan, at the southern extension of the spiral arm running through h and x Persei.

HD 64315 (m_v=9.3) is an O5 star in the

galactic cluster NGC 2467, which together with two other early O stars excites the extended H II region surrounding this cluster. An unpublished investigation of the colors and spectral types of the stars in and around this cluster, started at the McDonald Observatory, indicates that this star is at a distance of about 3 Kpc. The interstellar K line in HD 64315 appears double on 10 A/mm plates, both components being strong; the total intensity is of the order of 1 A.

Miscellaneous

The program on abundances in stars of the Pleiades and Hyades was seriously hindered by the bad winter weather, which prevented the taking of most of the coudé spectra required for this work. An interesting by-product of this program is the discovery by Deutsch of the only known double-line spectroscopic binary in the Pleiades, HD 23642. It has a range of nearly 200 km/sec, and a period of probably less than one day; its duplicity had been suspected by Struve and Smith on the basis of three discrepant velocities. HD 23950 proves to be another interesting member of the group, with abnormally strong lines of Mn II.

The open cluster IC 4665 was studied by Deutsch with spectra made at the 60-inch in an attempt to find the distribution of rotational velocities among the brightest stars. Struve had earlier found that among the late B stars in the Pleiades, the proportion of $V \sin i > 150 \text{ km/sec}$ is five or six

times greater than among noncluster stars of the same types. The spectra of 15 B stars in IC 4665 indicate that the distribution of rotational velocities there is like that in the Pleiades, and unlike that of noncluster B stars. A significant by-product of this investigation was the discovery of a well developed shell in HD 161261, a member of the cluster.

The spectrum of MWC 300, an interesting object of magnitude 10.5 that appears to be intermediate between ordinary Be stars and planetary nebulae, was photographed with a dispersion of 9 A/mm with the 200-inch telescope. Studies of this plate by Merrill indicate the interesting possibility of determining accurate wave lengths of numerous forbidden lines in various objects.

A striking peculiarity has been discovered by Greenstein in the rotation of the F and G giant stars. With only one exception, all A and F giants (luminosity class III) examined have shown considerable rotational broadening. Even some Go III stars show rotation. Apparently stars in the "Hertzsprung gap" have a strong tendency to rotate.

Single spectrograms of 9 stars in the nucleus of the globular cluster M 92 were obtained with the 8-inch camera of the coudé spectrograph of the Hale telescope and measured for radial velocity by O. C. Wilson. They show a range in velocity of 16 km/sec; the probable error of the individual plates is only 2 to 3 km/sec. This gives promise of yielding a definite value for the mass of this cluster.

NEBULAR RESEARCH WITH THE LARGE REFLECTORS

During the past year progress was made along many lines, including major research programs, exploratory work in new fields, and the development of instruments to exploit fully the great light-gathering power of the 200-inch telescope. The program of the distance scale has developed into a general study of type II stellar populations in globular clusters and extragalactic nebulae, and for the first time normal novae and variable stars are reported in the Ursa Major Cloud and the Virgo Cluster. The

first phase of the program should lead to a reliable, revised distance of M 31 and hence to the recalibration of individual distance indicators for near-by nebulae, and the second, to a reliable luminosity function of the nebulae themselves to serve as a statistical criterion of remote distances. Definitive, quantitative results must await the setting up of extremely faint photometric standards, and, in this connection, experimental photoelectric measures are reported down to mpg=21.8, considerably fainter than any previously achieved. As an example of exploratory studies may be mentioned the identifications of several of the extended radio sources that have been discovered during the past two years.

IDENTIFICATION OF EXTENDED RADIO SOURCES

Ever since the rapid development of radio astronomy after the last war, Baade and Minkowski have been in close cooperation with the active radio research centers in Australia and England in the attempts to identify the cosmic radio emitters with astronomical objects. This cooperation came about in a very natural way because one of the first objects suspected of being identical with a radio source was the Crab nebula, the remnant of a galactic supernova of A.D. 1054, which had been extensively investigated at the Mount Wilson Observatory just prior to the last war.

It is well known that the identification of radio sources with astronomical objects meets with very great difficulties because the accuracy with which the radio observers can locate the position of the emitters is rather low. Only for the few exceptionally strong radio sources may higher accuracy be reached by the use of interference methods, with an estimated uncertainty of ± 8 seconds of arc in right ascension and ± 40 seconds of arc in declination for the best case so far observed. Obviously

positions of even this accuracy would be quite inadequate for a positive identification if we knew that the emitter was a stellar object. About two years ago, however, the Australian radio group at Sydney obtained the first indications that a considerable percentage of the bright radio sources are not stars, but extended objects of measurable diameters. In this case a positive identification becomes possible if the astronomical object coincides with the radio sources within the latter's error of measurement, and if the radio diameter of the source agrees with the optical diameter of the object. In this manner it became possible during the past year to identify three of the outstanding radio sources of the sky, Puppis A (by Minkowski) and Cassiopeia A and Cygnus A (by Baade).

Puppis A

In the position of this object, with an extent of the order given by the radio observers, a mass of peculiar filaments has been found by Minkowski. Except for its size, this object resembles Cassiopeia A, which is discussed below. Spectrograms obtained with the nebular grating spectrograph at the 100-inch show that the random velocities of the filaments are of the order of ±50 km/sec; no systematic velocity of expansion is apparent. Filaments of a somewhat similar type are also found at the preceding end of the very large source Cygnus X. Most of the area covered by this object is heavily obscured, and the preceding end is in the most transparent area. It is rather unlikely that Cygnus X is connected with any of the bright galactic emission nebulosities in this part of the sky, which have an entirely different distribution.

CASSIOPEIA A AND CYGNUS A

In the fall of 1951 Baade made a search at the 200-inch for the two strongest radio

sources in the sky, Cassiopeia A and Cygnus A, for which Mr. F. Graham Smith, of the Cavendish Laboratory, had communicated his accurate new positions in advance of publication. It turned out that Cassiopeia A coincides with the center of a very remarkable emission nebula, about 5.4 minutes in diameter:

Smith's position of Cassiopeia A (1950.0): $23^{\text{h}} \, 21^{\text{m}} \, 12.0^{\text{s}} \pm 1^{\text{s}} + 58^{\circ} \, 32'.1 \pm 0'.7$ Center of emission nebula (1950.0): $23^{\text{h}} \, 21^{\text{m}} \, 12.0^{\text{s}} + 58^{\circ} \, 31'.5$

Subsequent measures at the Cavendish Laboratory confirmed that Cassiopeia A has a diameter which agrees with that of the emission nebula $(5.6 \pm 0.2 \text{ minutes of arc assuming it to be a uniformly radiating disk, or 5.0 minutes of arc if the shell is assumed to be optically thin). There seems to be no doubt but that the nebula is the radio source.$

The other strong radio source, Cygnus A, coincides with an extragalactic nebula which is the brightest member of a rich cluster of nebulae:

Smith's position of Cygnus A (1950.0): $19^{h} 57^{m} 45.3^{s} \pm 1^{s} + 40^{\circ} 35.0 \pm 1$ Position of extragalactic nebula (1950.0): $19^{h} 57^{m} 44.6^{s} + 40^{\circ} 35.7^{s}$

This nebula is a very queer object. In fact, its structure seems to make sense only if it is assumed that we are dealing with two nebulae which are in actual collision. If this interpretation is correct, the spectrum of the colliding gases in the two nebulae, an emission spectrum of high excitation, should be the outstanding feature. A spectrogram obtained by Minkowski with the nebular grating spectrograph at the 100inch revealed indeed a strong emission spectrum, with [Nev] as one of its features. That we are dealing with an extragalactic object is clearly indicated by the red shift of 16,500 km/sec of all emission lines. At first there was a contradiction between the diameter of the nebula, which is of the order of 30 to 45 seconds of arc, and the diameter of Cygnus A, for which the Cavendish observers had obtained the admittedly uncertain value of 3.5 minutes of arc. Later diameter measures at Manchester with a much longer base line, however, showed that the diameter of Cygnus A is of the order of 30 to 50 seconds of arc, in good agreement with that of the nebula. We have, therefore, every reason to believe that the identification of Cygnus A with the colliding nebulae is correct.

PLANETARY NEBULAE

Minkowski's study with the 100-inch of radial velocities of faint planetaries in the central region of the galactic system is progressing slowly, but he can now report that the dispersion of these velocities is of the same order of magnitude as that indicated by absorption lines in the central part of extragalactic nebulae. His study of the structure of planetaries based on monochromatic photographs with the 200-inch is nearing completion. During the year, some objects have been observed with bright central bodies of apparently spherical or nearly spherical symmetry but with faint outer parts which indicate that the conditions of growth of the gaseous envelopes are much more complicated than has been assumed hitherto. In certain cases, no axis of symmetry can be identified.

A substantial number of additional plates have been taken by O. C. Wilson for his study of the internal motions of the gases of the planetary nebulae. Plates of IC 418 taken with the 73-inch camera at Mount Wilson and the 144-inch camera at Palomar have resolved the [N II] lines $\lambda\lambda6548$, 6584 and the [O II] lines $\lambda\lambda3726$, 3728, all of which appeared single on previous plates of 10 A/mm. On the Palomar plate the [O III] lines remain sharp and

single even at 2.3 A/mm. This work is important in extending the kinematic results on this low-excitation nebula and in showing that those radiations [O III, Ne III] arising entirely in the central regions show very small expansion velocities, whereas those radiations showing a ring structure on slitless spectrograms yield definite and much larger expansion velocities under sufficient dispersion. Among the ring-type radiations the velocities correlate well with the excitations in the usual sense: the lower the excitation, the larger the velocity.

A 7-hour exposure of NGC 7027 by Bowen using the 8-inch camera of the 200-inch coudé brought out many more lines than those previously observed by Wyse. These included over a dozen members of the 5G-nH series of *He* II.

THE DISTANCE SCALE FOR EXTRA-GALACTIC NEBULAE

PHOTOELECTRIC STANDARDS

Priorities in the cosmological program were again assigned to the distance scale and to the law of red shifts. As was mentioned in last year's report, the program of the distance scale includes several projects, each a necessary step in the program, but also representing a contribution to other investigations of major importance.

The first step, the setting up of fundamental photoelectric standards extending to extremely faint limits in nine Selected Areas, has been continued by Baum. The present phase covers the range mpg=9 to 19, and more than 90 per cent of the selected stars have been measured at least once. In order to push the standards to extreme limits, a new 200-inch photometer is under construction, designed to implement special techniques developed for the task. The preliminary work included experimental measures of stars down to m_{pg}=21.8, well beyond limits previously reached, and laboratory tests which demonstrated the linearity of response of the

photo-tube down to the limit of measurability.

Color-Magnitude Diagrams of Globular Clusters

The second step in the distance program is the redetermination of distances of a few of the nearest globular clusters in high latitudes, in order to calibrate their brighter stars as distance indicators for near-by extragalactic nebulae. The distances will emerge from comparison of color-magnitude diagrams for the clusters with the similar diagram for stars in the neighborhood of the sun whose distances are known from direct triangulations. The definitive comparisons must be made at very faint absolute magnitudes, and, consequently, depend upon the first step in the program. Meanwhile, photoelectric sequences have been established in a few of the clusters, extending nearly to the desired limits, and photographic plates have been used to interpolate large numbers of stars into the standard system.

During the report year Sandage finished the measures of a set of Messier 3 plates taken at the 100-inch; Dr. Malcolm Savedoff finished a similar set of Messier 13 plates taken by Baade at the 200-inch. In both clusters the faintest photoelectric standards have to be extended by another 1.5 magnitudes in order to include the faintest stars measured on the present series of plates. Sandage's work on Messier 3, which is the most advanced at present, clearly shows that the giant branch in globular clusters continues through the subgiants and joins the main sequence at about F7. In both Messier 3 and Messier 13, blue stars of absolute magnitude o and fainter are frequent.

THE PERIOD-LUMINOSITY RELATION FOR CEPHEIDS OF POPULATION II

Since the recognition of two types of stellar population, it has become an open ques-

tion whether the cepheids of population II have the same period-luminosity relation as the classic cepheids (type I). Although the number of known long-period cepheids in globular clusters has been considerably increased in recent years, the available photometric data are of such poor quality that a period-luminosity relation based on them would hardly come up to standard. In order to provide better data, Arp has started a new determination of the lightcurves of long-period cepheids in all those globular clusters which are readily observable from Mount Wilson. The common zero point in all clusters will be represented by the mean brightness of the cluster-type variables. To ensure accurate photometric scales on the International System, the standard sequences in each cluster have been established photoelectrically by intercomparisons either with the pole or with one of the new primary Selected Areas. The final data will provide for each cepheid the photographic and the photovisual light-curve and, for the cluster-type variables of each cluster, their mean brightness on the International System.

Type II GIANTS IN GLOBULAR CLUSTERS

The study of the globular clusters has developed into a general study of the type II population, approached from various directions. Baum, for instance, who has made many photoelectric measures to calibrate the photographic measures made by others in M 3, 5, 13, 14, and 92, has extended his own sequences in M 3 and M 92, in order to compare the color-magnitude diagrams, as derived from the precise measures. The findings confirm those suggested by the earlier data, both photoelectric and photographic, namely that the red giants and yellow subgiants in M 3 are systematically redder than those in M 92; and Baum reports that the difference in the color index is about 0.17. The difference is similar to those found between sequences of blue giants in open clusters, and is believed to be significant from the viewpoint of chemical composition and stellar evolution.

Through the co-operation of Bowen and O. C. Wilson, spectra of 13 cluster stars were obtained with the new 8-inch camera of the 200-inch coudé, which has a dispersion of 38 A/mm. Not only did these spectra confirm the intrinsic difference found photoelectrically, but they also revealed that globular-cluster giants are quite different from any other known giants. The spectral type as determined by Baum for a typical M 92 giant having $M_v = -3$ is about G₃ on the basis of a criterion sensitive to excitation temperature, about F3 if judged from metallic lines, and about K₃ according to the color. The weakness of the metallic lines may be an exaggerated manifestation of the same weakness found in high-velocity stars. The surplus redness of cluster giants increases with luminosity and is somewhat similar to that found in ordinary giants of population I, except that in the latter case the surplus redness is a different function of luminosity and is less extreme.

Andromeda Nebula (M 31)

The third step in the distance program is the determination of the distance of M 31, using the distance indicators calibrated in the globular clusters. The result will serve to calibrate the brighter elements in the spiral, including cepheids, novae, etc., as distance indicators that can be recognized far out in space, and also will facilitate the general study of M 31 and its contents, as a stellar system curiously similar to the galactic system itself.

Baade, who has taken over the general supervision of this project as well as of the globular-cluster study, reports that the observations at the 200-inch of the three Andromeda fields mentioned in the last report were concluded at the end of 1951. The evaluation of the accumulated plate material started early in 1952 after Miss Swope joined the nebular department. Continued search for variables in the three fields has increased their numbers to more than 900, among them being 9 novae. Since the search is far from complete, it is evident that the 200-inch plate material collected during the two seasons 1950 and 1951 will provide a rich source of information regarding the variables in an Sb spiral. Miss Swope has nearly finished the light-curves for the field closest to the nucleus of the Andromeda nebula. They are at present given in readings of an arbitrary photometric scale, but are readily transformable into magnitudes on the International System as the final magnitudes become available for SA 68, with which the Andromeda field has been rigidly connected.

Among the results of general interest which are beginning to emerge, one is worth mentioning. It concerns the absorption effects which have their origin in a spiral like the Andromeda nebula. Up to now there has been a strong tendency to ignore the internal absorption in extragalactic nebulae, that is, to consider the cepheids in these systems as little or not at all affected by absorption. The results now available show conclusively that the cepheids of the Andromeda nebula are severely affected by absorption just as are the cepheids of our own Galaxy. Instead of clustering closely around the period-luminosity relation, the cepheids in the field closest to the nucleus of the Andromeda fill the whole area below the curve to the present plate limit, not a few of the cepheids indicating absorption of 2.5^m and more. Since the cepheids of this field are strongly concentrated in the spiral arms, their behavior is in agreement with a recent conclusion of Baade, based on other evidence, that the spiral arms are regions of abnormally high concentration of dust. It is hardly necessary to point out that the same arguments apply to the bright supergiants in such systems and that spurious results may be obtained if the presence of absorption is ignored.

NEBULAE BEYOND THE LOCAL GROUP

The fourth step consists in assembling distances of a large number of nebulae within the range of distance indicators to be calibrated in M 31. The list includes isolated nebulae and groups out to, and including, the Ursa Major Cloud and the Virgo Cluster. The total number should be sufficient to furnish a reliable luminosity function (the fifth step) which can be used as a statistical criterion of distance out to the limits of telescopes. Hubble has assumed responsibility for this phase of the program, and many plates have been contributed by almost every member of the nebular department of the Observatories. Progress was seriously hampered by bad weather during the winter and spring, and the report will be limited to two small groups, in addition to the Ursa Major Cloud and the Virgo Cluster.

THE M 81 GROUP OF NEBULAE

Results for the M 81 group, together with the neighboring wide pair NGC 2366/2403, 10 nebulae in all, now include 20 novae (19 in M 81 and 1 in M 82) and 25 variables (8 in M 81 and 17 in NGC 2403). One nova each was found by Humason, Baum, and Sandage; the other novae and the variables, by Hubble. Of the variables, 12 are definitely identified as cepheids with periods from 17 to 40 days, and light-curves have been derived, using an arbitrary scale of magnitudes. Several others appear to be cepheids with comparable periods, but no satisfactory curves have been constructed. When the faint photometric standards are available, reli-

able distances will be known in terms of M 31 as the unit, as well as dimensions, total luminosities, and luminosities of the brightest stars, for the entire group. To complete the study of this group, a project was initiated, in co-operation with Humason and Baade, for deriving masses of M 81 and NGC 2403 from spectrographic rotations measured in emission patches far out on the major axes of these spirals. Baade searched the nebulae for emission patches, using the method previously developed in the survey of M 31. Patches suitable for rotation measures were located in both spirals. In M 81, because of its resemblance to M 31, the search was extended to the main body as well, where more than 300 emission patches were found, concentrated, as in M 31, along the spiral arms. Bad weather restricted Humason's spectrographic observations to one plate each of two patches at intermediate distances on either side of the nucleus of NGC 2403. The preliminary data suggest that the mass-luminosity coefficient for 2403, a moderately faint Sc nebula, is comparable with that for the normal Sc nebula M 33.

M 101 GROUP OF NEBULAE

Fragmentary observations of the M 101 group produced one normal nova in the main spiral, but no additional variables. The bright star mentioned in last year's report was still faintly visible throughout the present season, and is now believed to be an irregular variable star rather than a supernova on the decline, as was at first suspected.

Normal Novae and Variables in the Ursa Major Cloud and the Virgo Cluster

The first normal novae and variables to be observed in the Ursa Major Cloud and the Virgo Cluster were identified during this season by Hubble. In spite of bad weather, enough 200-inch plates of NGC 4258, M 94, and M 87 were obtained to raise the total number of usable plates to about 8 to 10, including one red exposure, for each nebula. NGC 4258 and M 94 are giant Sb nebulae, the first definitely, and the second probably, a member of the Ursa Major Cloud. NGC 4258 produced 2 novae and 5 variables, while 1 nova and 6 variables were found in M 94. M 87, a supergiant elliptical nebula which is a prominent member of the Virgo Cluster, produced 2 probable novae and 3 variables.

GLOBULAR CLUSTERS AS DISTANCE INDICATORS

In the three Sb nebulae M 81, M 94, and NGC 4258, as well as the elliptical nebula M 87, semistellar images are found which, from their appearance, have been regarded as possibly globular clusters. Red exposures, although uncalibrated, now indicate that the colors of these objects are midway between the bluest (variables) and reddest (occasional stars and faint nebulae) objects recorded on the plates. Similar results have been found for NGC 3115 and 4594. Thus the colors of these objects, as well as their appearance and order of luminosity, are consistent with their interpretation as globular clusters, and they can be used as distance indicators, in particular for giant nebulae with type II populations. The criterion is welcome because the accumulation of normal novae in the Virgo Cluster and Ursa Major Cloud will proceed slowly even when the data for the two groups are combined with corrections for relative distance derived from the ratio of red shifts.

The calibration of clusters as distance indicators will be made most readily in M 31. With this end in view, Baum has already made photoelectric measures of integrated magnitudes and colors of about a dozen clusters in the great spiral, besides deter-

mining the radial distribution of light in some of the images from measures through graduated concentric apertures. The brightest known cluster is Hubble's no. 12, $m_{pg} = 14.98$, C.I. = 0.66 on the International Scale.

TOTAL MAGNITUDES OF NEBULAE

The fifth step in the distance program, namely the formulation of a luminosity function of nebulae, calls for a reliable system of nebular magnitudes. Because luminosities fade away from the centers outward to undefined boundaries, total magnitudes depend upon diameters (or apertures of measuring instruments), and, for photographic images, the diameters seem to increase with exposure up to the limit set by the sky background. Two problems are thus presented: the first is that of getting consistent, reliable measures for given diameters, and the second, that of assigning diameters (or apertures) which will ensure that the measures will exclude at most only negligible fractions of the total light.

Photoelectric measures, which are especially well adapted to the highly concentrated early-type nebulae, have been assembled with the Mount Wilson reflectors by Stebbins and Whitford, whose list of 152 objects was published during the past year, and more recently by Pettit, whose list of measures of 561 objects made during the past five years, and including most of the nebulae for which Humason has obtained spectra, has been prepared for publication. For many of the first list, and all of the second, colors were measured as well as total magnitudes. For 76 nebulae included in both lists, and measured with the same apertures, the average difference is P-SW = +0.005, and for 36 of these with measured color indices, the average difference in the color indices is P-SW = +0.03. Successive apertures were used for many

nebulae; in Pettit's list, for instance, three or more were used on 55 objects in addition to the small apertures with which nuclei or central condensations were measured in most of the nebulae.

These data may also be compared with the list of magnitudes (no colors) for 176 nebulae derived by Bigay, of the Observatories of Lyon and of Haute-Provence, from photographic plates using the Fabry method with, in general, series of apertures. For 67 nebulae measured by Pettit with the same apertures, the average difference is P-B=-0.06, with 5 residuals exceeding 0.5 mag., and 25 exceeding 0.2 mag. These large residuals, because the plus and minus signs are about equal, suggest accidental errors, and do not seriously affect the satisfactory agreement in zero points and scales. The comparisons indicate good progress in obtaining consistent magnitudes for given diameters.

Examination of the magnitudes for a given nebula measured with successive apertures (both the photoelectric results and, in particular, Bigay's list) emphasizes the desirability of devising some means of estimating over-all diameters on a consistent system. Otherwise it will be necessary to measure each object with a series of apertures and thus determine an asymptotic value of the total magnitude. Because the plates of the 48-inch schmidt telescope reach the sky background in practically all cases, and thus offer a rather uniform limit to exposures, a program has been initiated for making eye estimates of diameters of nebulae included in the three lists, and for running out profiles of as many as practicable with a registering microphotometer. The preliminary results suggest that it may be possible to calibrate eye estimates of diameters for different types, to assign apertures required for reliable measures of total magnitudes, and perhaps to derive corrections for improving incomplete measures.

Baum has made a contribution to the general subject by photoelectric measures, through series of apertures, of magnitudes and colors of the bright globular nebulae NGC 3379 and 4552. From these measures he has derived profiles of the general form indicated by earlier photographic investigations, but on a quantitative and much more reliable basis, especially in the outer regions. He concludes that an appreciable fraction of the total light of these nebulae originates in outer regions so tenuous that the eye cannot detect them on photographs. He reports further that the nuclei are slightly redder than the outer parts $(\Delta C.I.\sim 0.07)$, as, he points out, is the case in globular clusters.

THE LAW OF RED SHIFTS

Effects of Red Shifts on Apparent Magnitudes

The formulation of the law of red shifts involves not only determination of magnitudes and red shifts, but also reliable estimates of effects of red shifts on magnitudes. Energy-distribution curves (Stebbins and Whitford) for spectra of E, Sb, and Sc nebulae now permit direct estimates of the corrections for various colors, using E nebulae alone for the great clusters, and the observed mixture of types among bright (Shapley-Ames) nebulae for the general fields. Corrections to photographic magnitudes ($\lambda = 4320$) involve extrapolations beyond $d\lambda/\lambda = 0.2$, but, subject to this uncertainty, Hubble has found that they are well represented by the relations $\Delta m =$ 3.od λ/λ and 3.6d λ/λ , for field and cluster nebulae respectively, out to $d\lambda/\lambda = 0.4$ or more. These results fully confirm the values, 3dλ/λ, used in previous discussions of the distribution of nebulae in depth, but introduce revisions in the data used for the law of red shifts, increasing up to a maximum of 0.08 mag., for the Boötes Cluster.

Corrections to photovisual magnitudes $(\lambda=5290)$ are numerically smaller and involve no extrapolations out to $d\lambda/\lambda=0.4$ or more. They are not linear functions of $d\lambda/\lambda$, but can be represented by linear segments in successive ranges of $d\lambda/\lambda$. Thus for clusters, the corrections out to $d\lambda/\lambda=0.1$ are $\Delta m=1.7d\lambda/\lambda$, and beyond that, out to $d\lambda/\lambda=0.4$, they are $\Delta m=2.86d\lambda/\lambda-0.11$. Corrections for field nebulae are considerably smaller.

Law of Red Shifts Derived from Photovisual Magnitudes

These corrections have been used in reformulating the law of red shifts on the basis of photovisual magnitudes in 12 clusters either measured directly with phototubes or derived from photographic measures calibrated by photoelectric measures (Whitford, Stebbins and Whitford, Pettit). The results—which represent an observed range of red shifts out to 0.2, or 50 per cent greater than that previously used, reliable corrections for red shifts involving no extrapolations, and much improved photometric data—are about the same as the earlier formulations based on photographic magnitudes. If recession factors are ignored, the law remains sensibly linear. If recession factors are included, the law departs from linearity in the sense that, when it is expressed as a power series of the distance, $d\lambda/\lambda = kr + lr^2$, the coefficient l is positive. Relative distances may be expressed in terms of the Virgo Cluster (or any of the others) as a unit. Using the Virgo Cluster, and assigning it a distance of 7×10^6 light years, the values of k and l are $+5.4 \times 10^{-10}$ and $+5.3 \times 10^{-19}$, not very different from those previously derived.

Programs for Testing the Linearity of the Law of Red Shifts

Expected revisions of the unit of distance will change the numerical values of k and

l, but not the sign of *l*. This term, if it is not real, must reflect systematic errors in the magnitude scale, or estimated corrections for red shifts, or a secular (evolutional) fading of the giant elliptical nebulae observed in the clusters. Programs have been formulated to test all three possibilities.

Because corrections for red shifts in E nebulae are a minimum for red magnitudes (at $\lambda = 6300$ they remain less than ± 0.1 mag. out to $d\lambda/\lambda = 0.3$), a program is under way to obtain such magnitudes of the dozen or so brightest nebulae in each of the clusters used in formulating the law. For this purpose a jiggle camera has been constructed for use at the prime focus of the 200-inch. Magnitudes in the blue and yellow, obtained with this camera, will be tested and calibrated by photoelectric measures, and thereafter the camera will be used not only in the blue and yellow, but also in the red, where the photo-tubes are not now sufficiently sensitive. Baum has already made special measures of nebulae in the Corona Borealis Cluster to facilitate the calibration.

The possibility of secular fading will be tested by assembling supernovae in distant clusters to compare the mean distances of the group of clusters as indicated by the supernovae and by the apparent magnitudes of the brighter nebulae. Preparation of a list of suitable clusters was initiated during the year, but was deferred until a broader basis for an optimum selection

should be furnished by survey plates with the 48-inch schmidt telescope.

VELOCITY DISPERSIONS IN THE COMA AND THE CORONA BOREALIS CLUSTERS

Although Humason's attempts to record spectra of very distant nebulae were completely stopped by bad weather and poor seeing, he was able to assemble a few spectrograms of relatively bright nebulae in the Coma and Corona Borealis clusters. In Coma, he obtained improved spectra of 6 objects previously known from poor plates, and first spectra of two nebulae about 1° from the center of the cluster. The new material does not substantially alter the previous values of the mean red shift and the dispersion (v = +6670 km/sec and $\sigma = 1050$ km/sec).

In Corona Borealis, 7 nebulae were newly observed, to make a total of 11 spectra of 8 different nebulae. The mean red shift (uncorrected for galactic rotation) and the dispersion are v=+21,530 km/sec and $\sigma=1210$ km/sec. The dispersion is dominated, however, by two very large residuals, one plus and the other minus, and if these two measures are ignored, the mean red shift is sensibly unchanged while the dispersion is cut nearly in half. For this reason, it is believed that the suggestion that dispersions may increase with distance should not be given much weight at this time.

RESEARCH WITH THE SCHMIDT REFLECTORS

THE NATIONAL GEOGRAPHIC SOCIETY— PALOMAR OBSERVATORY SKY SURVEY

The National Geographic Society-Palomar Observatory Sky Survey was carried on by A. G. Wilson and R. G. Harrington during the 95 nights of the year in which at least some of the hours were suitable for observations. About 60 per cent of the sur-

vey fields have now been photographed, but 35 per cent of these plates will be repeated in order to achieve the uniformly high standard set for acceptance.

Three comets were found on the survey plates (1951i and k, and 1952a, the second being a rediscovery by Harrington of Wolf 2, found in 1924). Among the many

asteroids recorded, one (1951RA) was found with a perihelion distance of 0.827 astronomical units.

Preliminary inspection of the plates continues to show large numbers of new objects which emphasize the significance of the survey. These objects include large faint planetary nebulae (one 13' in diameter), faint clouds (galactic nebulosities) in galactic latitudes as high as 75°, unusual star clusters, and, of course, many clusters of nebulae from which excellent examples can be selected for further investigations of red shifts.

NOVAE IN THE GALACTIC SYSTEM

Zwicky found two novae during his search of selected regions of the Milky Way, using the 18-inch schmidt with an objective prism. The objects were:

	Nova in	Nova in
	Aquila	Cygnus
Discovery date	July 10, 1951	Sept. 29, 1951
R.A. 1950	19 ^h 5 ^m 17 ^s	17 ^h 50 ^m 0 ^s
Dec. 1950	+10° 25′.8	-29° 33′.8
Apparent mag	+ 11.5	+ 12.5

FAINT BLUE STARS AND VARIABLES NEAR THE GALACTIC POLES

Zwicky has extended his search for faint blue stars in high galactic latitudes, beyond the limit ($m_{pg}=15$) previously reached, nearly to $m_{pg}=20$. He has also started an investigation of faint variables near the galactic poles. Most of the stars were found in his extensive search for supernovae with the 18-inch schmidt, but the program has now been extended with the 100-inch reflector to include stars as faint as $m_{pg}=20$. The purpose, in both programs, is to investigate the possibility of a transition from the stellar population of the galactic system to a stellar population scattered through extragalactic space between the nebulae.

One of the brightest of the variables (R.A.=12^h 28^m 26^s, Dec.=12° 35'.1, 1950),

found independently by Zwicky although long known on Mount Wilson plates as a foreground variable in the field of M 87, 5' north preceding M 87, was followed by Zwicky and Sandage and found to be a long-period variable (P=180 days, mpg [max]=13.8). Dr. N. U. Mayall, who obtained a spectrum shortly after maximum, described it as that of a red giant.

DISTRIBUTION OF NEBULAE

Zwicky reports that he has counted more than a million nebulae on 48-inch red plates centered in Coma, Cancer, Pegasus, and Corona Borealis. Numbers per plate range from 100,000 (i.e., about 2500 per square degree) in Corona Borealis to about 20,000 in the region of the Coma Cluster. Analysis of the fluctuations indicates conspicuous departures from random distribution, and work is in progress to determine to what extent the nonuniformity is real, and how much must be referred to obscuration either within the galactic system or in internebular space. In this connection, unusually low counts of very faint nebulae in the vicinity of large clusters are presented as evidence for the concentration of internebular dust in these clusters. In collaboration with Wild, contours of "equal population" have already been constructed for the Shapley-Ames nebulae, and similar curves will be made for counts to the limit of the 18-inch schmidt which are now in progress.

Survey of Nebulae for Photographic Magnitude 15

Meanwhile steady progress is reported on the survey to the limit $m_{pg}=15$, as determined from images 1 mm square made with the 18-inch by schraffier methods. This program is being carried on by Zwicky in collaboration with Wild and H. S. Gates on an ONR contract. The goal is a general catalogue listing positions and magnitudes

of the nebulae, $m \le 15$, within reach (order of 15,000), and efforts are now concentrated on the belt between $Dec.=-4^{\circ}$ and $+24^{\circ}$ running from R.A.= 7^{h} to 16^{h} . The data will furnish another set of equal-population contours.

Luminous Material Between Nebulae

On both 48-inch and 18-inch plates Zwicky has assembled a considerable number of cases of faintly luminous filaments and clouds which connect or lie between components of double or multiple nebulae, as well as groups and clusters. In many of these cases, the separation of components is much wider than any known before the schmidt plates were available. A few cases have been selected for detailed study with the large reflectors; for example, a remarkable triple nebula in Virgo whose components are IC 3481 (Sop), IC 3483 (Sap), and an anonymous Sop nebula between them.

Analysis of Clusters

Analysis of clusters of nebulae has been continued by Zwicky in order to find

whether or not his "structural indices" vary with distance—a question which may bear on the time scale of the universe. A new approach to the problem of the time scale is believed to have been found in the frequency distribution of angular diameters of clusters, which, for distances greater than about 2×108 light years, should differ materially, he believes, according to whether or not the universe is expanding at the rate suggested by the red shifts. This possibility has led to a detailed examination of clusters on some of the 48-inch plates. On one plate, in Corona Borealis, he identified about 100 clusters (more than 2 per square degree), and another observer has reported other regions of high frequency between Coma and Virgo. The data, together with an observed sparsity of distant clusters in regions covered by large, near-by clusters, are considered to furnish additional evidence of the existence of internebular material both nonuniformly scattered in space and concentrated in the clusters. Some of the faint clusters found with the 48-inch and 100-inch are believed to be more distant than any reported in the literature so far.

THEORETICAL STUDIES

Greenstein and Minkowski have investigated the theory of the radio-frequency emission of the Crab nebula, which coincides with radio source Taurus A. The latter has an unusual radio-frequency energy distribution, essentially flat from 30 to 1210 mc/s. The energy in other discrete radio sources varies inversely as the frequency, as it does in many of the disturbed outer regions of the sun. New models of the Crab nebula with high electron temperature have been computed to explain the visual continuous spectrum, but these fail to predict the radio-frequency emission. Thus the Crab nebula must include

nonthermal radio generating sources. The opacity due to free-free transitions in the electron-ion cloud for all possible models is found to be appreciable at 100 mc/s. If ordinary nonthermal sources are located inside an absorbing cloud, the predicted spectrum has a flat maximum, in agreement with observation. These considerations may also apply to some of the other gaseous radio sources identified by Baade and Minkowski.

The distribution of color excesses of early-type stars has been investigated from a statistical standpoint by Münch. The observational data used are the color meas-

ures of Stebbins, Huffer, and Whitford and unpublished determinations of intrinsic colors and spectral types by W. W. Morgan. On the basis of Ambarzumian's discrete-cloud hypothesis, an attempt has been made to determine, from the observed color excesses of stars in a certain range, the mean, the variance, and the skewness of the absorbing powers of the units which are supposed to build up the interstellar medium. The inadequacy of Ambarzumian's model to describe in any more detail the observed characteristics of the color-excess distribution has been discussed.

Paper III of the series "On the Theory of the Brightness Fluctuations of the Milky Way," published by Chandrasekhar and

Münch, raised a problem of some mathematical interest which was left unsolved. The surface brightness u, in a finite system of uniformly distributed stars and randomly distributed absorbing clouds, is given as a stochastic integral, along the line of sight, of the product of the absorbing powers of the intervening clouds. It is known that the probability distribution function of the brightness, $f(u,\xi)$, satisfies certain differential equations. The problem is to show that, when all clouds are supposed to have the same optical thickness, the differential equation completely determines $f(u,\xi)$. Recently Münch found the solution to this problem, which has some novel mathematical features.

INSTRUMENTATION

Two important additions were made to the spectroscopic equipment of the 200inch Hale telescope during the current year. The first of these was a new spectrograph body for the nebular prime-focus spectrograph. This is designed to use a grating instead of the prisms of the old instrument. Two gratings of 300 and 600 lines per millimeter, each blazed in the second order, are being provided. The old thick-mirror schmidt camera of 1.4-inch focus and 3-inch aperture of the prism camera is used with the new grating instrument, and in addition a 2.85-inch-focus camera of the same type is being provided. The various combinations of gratings and cameras yield dispersions from 110 to 440 A/mm.

The second addition during the present year was the completion of the cameras for the medium dispersions of the coudé spectrograph. This included the final figuring of the corrector plate for the 18-inch camera and the completion of the quartz aplanatic sphere and its mounting for the 8.4-inch camera. The first of these cameras gives a

dispersion of 18 A/mm in the third-order blue and 27 A/mm in the second-order red. The corresponding dispersions for the 8.4-inch camera are 38 and 57 A/mm. The tests indicate that these cameras give critical definition. Because of their extremely short focal ratio (F/D=1.5 and 0.7, respectively), they are capable of reaching the very faint magnitudes of 13 and 15, respectively, with a 6- or 8-hour exposure under average seeing conditions.

Swanson, working under the direction of H. W. Babcock, has ruled twenty gratings with the "B" (newer) ruling engine during the year. Two were for the prime-focus nebular spectrograph, two for the new 60-inch universal spectrograph, three were for a calibration instrument, and one is a concave. Some experimental gratings were made having 900 grooves per millimeter and maximum intensity in the third order. Three gratings were made having a ruled area of 5 by 8 inches. The best of these is blazed in the fifth order, and in the 75-foot spectrograph of the Hale Solar Laboratory is providing a dispersion of 1A

per 11 mm and a measured resolving power of 600,000. Scattered light and ghosts are negligible. Such a grating makes possible a new approach to several problems in solar physics.

With the "A" ruling machine, several experimental fairly coarse transmission gratings have been ruled directly on plastic sheets. The largest is 18 inches square. These gratings show considerable promise if better-quality plastic can be obtained.

For some years, H. D. Babcock has been working on the problem of a general magnetic field of the sun with an interferometer and a concave-grating spectrograph. This equipment has now been superseded by the new fifth-order plane grating, men-

tioned above, that has been installed in the 75-foot spectrograph of the Hale Solar Laboratory. In collaboration with H. W. Babcock, a new photoelectric detector for the Zeeman effect has been designed and built. Recording equipment, in the form of a direct-coupled cathode-ray tube and a camera, has been provided. With this apparatus, magnetic activity over the whole disk of the sun is now being mapped daily, using a scanning technique. Areas of persistent magnetic activity in the vicinity of sunspot groups are easily revealed and can be followed from day to day. Other and weaker phenomena are also under investigation.

GUEST INVESTIGATORS

The Observatories have continued to invite a substantial number of guest investigators from other institutions to make use of their observational facilities and their files of plates. The following investigations have been carried out during the current year.

Dr. William P. Bidelman, of the Yerkes Observatory, spent several weeks in the summer of 1951 studying spectrograms in the files to obtain information for a catalogue of late-type stars showing emission lines.

Observations of comets, asteroids, and satellites were made during nine months of the current year by Dr. Leland E. Cunningham, of the University of California. Observations included the following objects which were discovered at these Observatories: 1952a P/Harrington-Wilson, 1951k P/Wolf-Harrington, 1951i Comet Wilson-Harrington, 1950b Comet Minkowski, 1566 Icarus, 1951RA Wilson-Minkowski Object, 1950LA Wilson-Wallenquist Object, and Jupiter satellites IX, X, XI, and XII. In addition, plates were obtained as often as practicable of 16 current

comets, 10 unusual asteroids, and other miscellaneous objects. A total of 464 plates were exposed for this program.

Miss Guro Giellestad, of the Institute of Theoretical Astrophysics, The University, Blindern, Oslo, Norway, spent nine months at the Observatories with the aid of a grant provided by the American Association of University Women. She investigated the reversing magnetic field of the peculiar Atype star HD 153882, using 20 spectrograms taken in 1948 to 1952 by H. W. Babcock with the double analyzer for circularly polarized light. The effective magnetic field intensity at the pole, H_p, as determined from measures of the Zeeman effect on about 30 lines on each plate, varies periodically between the limits +4500 gauss and -4000 gauss. The derived period is 6.005 days. The mean-velocity curve has a range of 5.3 km/sec, but systematic differences between elements are small. This finding is probably related to the fact that the spectrum shows little or no variability in line intensities. The spectrum is peculiar in that Cr I, II and Mn I are strong and the K line is sharp, but the star is not

a spectrum variable. A cross-over effect in the polarized line profiles, rather similar to that previously found in HD 125248 and α^2 Canum Venaticorum, was observed at phases when the effective magnetic field is near zero.

After discussion with H. W. Babcock and a study of his observational material on magnetic stars, Miss Gjellestad also revised and completed for publication her studies of a theoretical model of a magnetic star which she had previously developed in Norway.

Using the coudé spectrograph of the 100inch telescope on Mount Wilson, Dr. George H. Herbig, of Lick Observatory, obtained spectra of T Tauri, RY Tauri, UX Tauri, SU Aurigae, and FU Orionis. These stars were chosen as being suitable for study of the absorption spectrum, which in such variables as RW Aurigae is nearly obliterated by strong emission. It is found that the spectra of these five stars are unusual in that they are all characterized by broad, diffuse absorption lines such as are produced by rotation or turbulence. Plates of a number of normal stars were also obtained for comparison with the bright-line objects. Four spectrograms of the cepheid variable S Sagittae, which last year was found to have emission in the H and K lines while on the rising branch of its light-curve, were also obtained.

From July 1951 to January 1952, Dr. Erik Holmberg, of Lund Observatory, continued his previous photometric program on near-by extragalactic nebulae. The work has aimed at a determination of total photographic and photovisual magnitudes of all those nebulae in the northern sky which, on account of their size, cannot be suitably observed by photoelectric methods. A total of about 500 plates were taken with the 60-inch telescope and 200 with the 18-inch schmidt instrument. All nebular exposures, which were made in focus, were calibrated by means of extrafocal exposures of polar

stars. The plates, which are being measured in Lund, will be used as the basis for a determination not only of integrated magnitudes, but also of distribution of luminosity and color in the nebulae.

During the winter of 1952, 102 lowdispersion spectrograms of S-type stars (and M-type comparison stars) were taken by Dr. Philip C. Keenan, of the Perkins Observatory, with the Cassegrain spectrographs on the 60-inch and 100-inch reflectors. These plates were taken to permit estimates of intensities of strong lines and bands in the region from 4300 A to 6000 A. The intensity estimates will help in the problem of ordering the S-type stars in a temperature sequence. Coudé spectrograms of R Geminorum (Se), R Hydrae (M7e), and R Leonis (M7e), taken by Bowen and Merrill, were measured in the yellow and red regions, since the strongest bands in this part of the spectrum have never been completely identified or analyzed. In the M-type stars the Dunér II and Dunér III bands were found to be identical with the system which Coheur produced at Liége by exploding titanium wires. The behavior of these bands, and their vibrational constants, suggest that they may arise from intercombination transitions between the normal level and a higher singlet level of the TiO molecule. In the S-type star R Geminorum, nearly all the strong bands are due to known systems of ZrO and YO, except for the very prominent head measured at 5849 A, which remains unidentified.

Dr. R. B. Leighton, of the Department of Physics of the California Institute of Technology, has continued his experiments with motion pictures of Jupiter. Two allnight runs have been obtained at the 60-inch telescope, using 16-mm Kodachrome film. The exposure duration on these runs was 1.5 seconds per frame with a lapse of 30 seconds between frames. One of these runs shows good detail and good color

rendition of the planetary disk. As was to be expected, the Jupiter films showed much greater detail when projected as a motion picture than when viewed frame by frame, the resolution in the former case being limited almost entirely by the mirror shape and the seeing.

The co-operative program with the Mc-Math-Hulbert Observatory for the direct recording of the solar spectrum was continued from July 1, 1951, to June 30, 1952, as in the preceding two years. Between July 1 and July 31 a new map of the solar spectrum from 0.8 to 3.7 microns was completed. For the wave-length range from 0.8 to 2.6 microns the sensitivity of the equipment has been improved so that the spectrometer normally operates with slit widths about 30 per cent narrower than the optimum predicted on the basis of the Rayleigh criterion. For wave lengths longer than 2.8 microns the spectrometer slits must be about 25 per cent wider than the optimum value, because of lack of sensitivity.

From July 31 to August 25, tracings for use in wave-length measurement and tracings of the sun's east and west limbs for the segregation of solar and terrestrial lines were made. Occasional re-runs for the improvement of atlas tracings were made during this interval. From August 24 to September 10, the limb tracings of the λ10830 line of helium and numerous other solar lines were made, to record the variation of line intensity from center to limb of some of the more conspicuously variable lines. On September 1, Mr. William Livingston started work as observer, replacing Mr. Dale Vrabec. From September 10 to April 5, the bulk of the observing time was spent in making east-limb, center, and west-limb tracings for studying changes in solar-line intensities from the center to the edge of the limb, and to continue detection of solar lines by the velocity shifts at the limb of the sun. On some of the limb tracings made in mid-October, solar lines of *CO* were found by Dr. Leo Goldberg.

Beginning on April 8, Dr. Keith Pierce, of the McMath-Hulbert Observatory staff, and Mr. Herbert E. Sanderson, instrument maker at the McMath-Hulbert Observatory, installed equipment to be used in measuring intensities in the solar continuous spectrum. This equipment consists chiefly of calibrated light sources to be compared directly with the sun. The installation was completed by April 15, and Mr. Sanderson left Mount Wilson at that time. From April 15 until June 30 the spectrometer has been running full time on solar-intensity measures.

Dr. Guy C. Omer, Jr., of the University of Chicago, continued his survey of the Coma Cluster of nebulae. This included extensive nebular counts to give the distribution of nebulae over the cluster, and an attempt to obtain the luminosity function of the nebulae in this cluster.

Dr. J. H. Oort, of the Observatory of Leiden, took a series of second-epoch plates for the study of the proper motions of RR Lyrae type stars.

Spectrograms were obtained by Dr.N.U. Mayall, of the Lick Observatory, with the Newtonian grating spectrograph of the 100-inch reflector, of 10 emission-line objects in M 31 for study of internal motions in the spiral, and of 13 galactic planetaries for a check of systematic errors. These 1951 plates, and those obtained in 1950, provide radial-velocity data in M 31 for 7 more faint patches and one stellar object, and for 8 patches for which velocities have already been published. With these new Mount Wilson data, it should be possible to derive a detailed rotational-velocity curve for M 31 from the velocities of 40 bright-line objects in the spiral.

Dr. D. M. Popper, of the University of California at Los Angeles, and Dr. E. C.

Yowell, of the National Bureau of Standards at Los Angeles, have obtained additional spectra of dwarf stars of types G to M having H and K in emission. Visual inspection of the spectrograms of the 6 dwarf stars indicates that there are no well marked changes in the intensity of H and K emission in any of them, although slight changes are suspected in two cases. If the negative results are borne out by a continuation of the observations, the conclusion will be that either H and K emission in the integrated light of the dwarf stars is not associated with prominence-like activity, or the amount of such activity does not fluctuate markedly.

Dr. Malcolm P. Savedoff spent the current year at the Observatories on a fellowship provided by the National Research Council. The profiles of the weak absorption lines in the spectrum of RR Lyrae were investigated in order to determine the applicability of the pulsation theory to this object. Additional plates were obtained of RR Lyrae and of several Fo and F2 comparison stars for spectrophotometric analysis of the atmospheres.

Savedoff has also made a study of the color-magnitude array of the globular cluster M 13. About 1400 stars, mostly between visual magnitudes 14 and 19, have been measured on plates taken by Baade with the 200-inch telescope. Reductions are still in progress. A moderate population at the faint blue end of the cluster variable sequence is noted, including stars fainter than visual magnitude +3.5.

Two spectroscopic programs on red giants were undertaken by Dr. Martin Schwarzschild, of Princeton Observatory. The purpose of the first program was a further quantitative investigation of the composition differences between the two stellar populations. Seven low-velocity and four high-velocity K giants were included. The wave-length region covered is λ4120

to λ 4460. In the second program, an attempt was made to determine the abundance of C^{13} (or an upper limit to this abundance) for normal giants with the help of the CN band at λ 3883. Twenty-eight G and K giants were put on this program. The spectra for both programs were obtained with the coudé spectrograph of the 100-inch telescope. The spectra have been registered and the reductions are now under way. Dr. Schwarzschild also collaborated with Richardson in a study of sunspot motions.

Dr. Otto Struve, of the University of California, has made an extensive study of stars of the β Canis Majoris and β Cephei type. Several hundred spectrograms were obtained with the 60-inch telescope of two representatives of this group, 16 Lacertae and v Eridani. For 16 Lacertae a series of plates were also secured at the coudé focus of the 100-inch telescope. These high-dispersion plates indicate that the lines are variable in profile and intensity. Both stars demonstrate strikingly the presence of two closely similar periods, the values in 16 Lacertae being 4^h 3^m and 4^h 6^m. This star has still another period of 12 days, which may be caused by ordinary binary motion.

Another group of plates was taken by Struve to study small spectroscopic differences occurring in stars in the solar neighborhood that lie close to each other in the Hertzsprung-Russell diagram. Examples of such pairs or groups of stars are β Comae Berenices and β Canum Venaticorum, and λ Aurigae and λ Serpentis. The purpose of this study is to ascertain whether the small differences in absorption lines of such stars can be fully explained in terms of classic ionization theory, or whether abundance differences and perhaps unknown effects complicate the picture.

Sister Mary Therese, of Mundelein Col-

lege, spent the summer of 1951 co-operating with Baade in tests of the photometric procedures which were later adopted for the Andromeda program.

Dr. A. E. Whitford, of the Washburn Observatory of the University of Wisconsin, used an improved red-sensitive photoelectric photometer and filter system in the summer of 1952 to study the colors of elliptical nebulae with a large red shift. Preliminary results confirm the 1951 conclusion that in the red and near infrared these nebulae do not show the color excess previously found for the same objects on the International System. The interpretation depends on the complete multicolor study extending down into the ultraviolet. The 1951 results on this part of the spectrum showed inconsistencies, and more extensive observations are needed.

Dr. A. D. Code, also of Washburn Observatory, made a study of the two-hour cluster variable EH Librae (BD -0°2911), based on two-color light-curve and radial-

velocity plates obtained in the summer of 1951. The data are consistent with the pulsation hypothesis, and the star appears to follow the same pattern as a classic cepheid. Spectrographic observations of the brightest stars in the open cluster NGC 6604 yielded data for tentative distance modulus, but spectra of more members are needed for a final conclusion.

Preliminary tests of a photoelectric spectrum scanner carried out by Dr. Code in the summer of 1951 were very encouraging, and led to the design and construction of a more versatile mechanical arrangement for use on a grating spectrograph. Stars down to the eighth magnitude can be scanned with a resolving power of 5 to 10 A, and the range from 3100 A to 10,000 A can be covered.

The photoelectric color survey of the O and B stars on the Nassau-Morgan list, carried on by Dr. Code and Dr. Whitford in the summers of 1951 and 1952, is nearing completion.

STAFF AND ORGANIZATION

During the report year two additions have been made to the personnel of the Observatories. Dr. Guido Münch came as staff member on July 1, 1951, and Henrietta H. Swope as research assistant on February 1, 1952. Dr. Alexander Pogo took over the duties of Editor from Dr. Paul Merrill on January 1, 1952.

RESEARCH DIVISION

Solar Physics: Seth B. Nicholson, Edison Pettit, Robert S. Richardson, Thomas A. Cragg, Joseph O. Hickox, James M. Parker. Stellar Spectroscopy: Paul W. Merrill, Horace W. Babcock, Ira S. Bowen, Armin J. Deutsch, Jesse L. Greenstein, Rudolph L. Minkowski, Guido Münch, Olin C. Wilson, M. K. Vainu Bappu, William Buscombe, Helmut Abt, Sylvia Burd, Mary F. Coffeen, Dorothy S. Deutsch, A. Louise Lowen.

Nebular Photography, Photometry, and Spectroscopy: Edwin P. Hubble, Walter Baade, William A. Baum, Milton L. Humason, Rudolph L. Minkowski, Edison Pettit, Albert G. Wilson, Fritz Zwicky, Halton C. Arp, Alice S. Beach, Allan R. Sandage, Henrietta H. Swope, Paul Wild.

Secretary of the Observatory: Milton L. Humason.

Editor and Librarian: Alexander Pogo. Photographer: William C. Miller.

Instrument Design and Construction

Design: Bruce Rule, project engineer; Edgar C. Nichols, chief designer; Harold S. Kinney, draftsman.

Optical Shop: Don O. Hendrix, superintendent; Floyd E. Day, Melvin W. Johnson, opticians.

Instrument Shop: Albert T. McIntire, superintendent; Oscar Swanson, instrument maker; Albert Labrow, Fred Scherff, Murdock McKenzie, Donald W. Yeager, machinists.

Maintenance and Operation

Mount Wilson Observatory and Offices

Office: Anne McConnell, administrative assistant; Wilma J. Berkebile, secretary; Leah M. Mutschler, stenographer and telephone operator.

Operation: Ashel N. Beebe, superintendent of construction; Hugh T. Couch, carpenter; Kenneth E. DeHuff, engineer; John E. Shirey, janitor and relief engineer; Eugene L. Hancock, Alfred H. Olmstead, Arnold T. Ratzlaff, night assistants;

Emerson W. Hartong, truck driver; Therese Ratzlaff, Evelyn J. Blake, stewardesses; Arthur L. France, Everett W. McConnell, janitors.

Palomar Observatory and Robinson Laboratory

Office: Eleanor C. Ellison, secretary and librarian; Dorothea Davis, secretary.

Operation: Byron Hill, superintendent, Palomar Observatory; Benjamin B. Traxler, electrician and chief night assistant; Charles Kearns, Robert E. Sears, night assistants; Harley C. Marshall, office manager; Ferd Feryan, mechanic; Gladys Feryan, stewardess; Ray White, powerhouse operator; George W. Pettit, janitor; Gus Weber, assistant mechanic.

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GEOPHYSICAL LABORATORY

Washington, District of Columbia LEASON H. ADAMS, Director

Progress of the newly oriented postwar program was reviewed in last year's report and found to be advancing at a very encouraging rate, mainly as a result of the new experimental techniques and equipment developed since the war. It is interesting to note that the foremost section of the program, namely the investigation of silicate melts in the presence of volatile materials at elevated pressures and temperatures, is essentially one of the four projects especially recommended by Dr. C. R. Van Hise in his report of 1902-1903 (printed in full in Year Book No. 2) on the advisability of establishing a geophysical laboratory. Van Hise wrote, in part:

Experiments should be carried on with aqueous solutions under various pressures and at various temperatures. The higher temperatures should approach those of magmas, in order that the relations of crystallization from magmas and crystallization from water may be learned. . . . It is certain that the conditions under which many of the minerals are produced in nature from water solutions can be produced in the laboratory. Only when this is done shall we have an adequate basis upon which to judge of the kinds of minerals that are produced in nature from aqueous solutions and their manner of formation.

It is also interesting to note that twenty years after the founding of the Laboratory the Director reviewed briefly the progress of the researches and reported little advance in the experimentation on the behavior of silicates in the presence of volatile materials. In commenting on this aspect of the program, Dr. Day wrote (Year Book No. 26, 1926–1927):

Although this [the past] program has proved to be sound and has been successfully

followed, it does not yet yield exactly the conditions of formation of the igneous rocks, for we know, also from laboratory experience, that all igneous rocks contain volcanic gases volatile ingredients such as water, carbon dioxide, sulphur compounds and halogens, which are now known to influence these conditions of formation radically and vitally. Indeed these volatile ingredients are chemically the most active components associated with the formation process. To accomplish this further step of determining precisely the role of the volatile ingredients in rock formation involves placing the whole process under such pressure as to hold the volatile ingredients confined throughout the operations studied. These pressures may be very high and the pressure technic is therefore certain to be enormously more difficult than ordinary laboratory procedure at extreme temperature under atmospheric pressure alone. In terms of engineering practice this means gas-tight bombs within which the appropriate temperature and pressure, high or low, may be held constant at any point through an enormous range in order that the proportions of the components participating in the formation process may not be variable during its operation.

The preliminary technical experience leading in this direction has been successfully attained, but the determination of the major mineral relations with appropriate volatile ingredients present still remains one of the foremost problems confronting us. It is also the first occasion in the course of our progress where the available laboratory equipment is not adequate for continuing the work on an appropriate scale. It is easily possible to carry out simultaneous experiments in a considerable number of open crucibles. It is a problem of quite another order of magnitude to provide strictly gas-tight bombs for a like number of studies, or presses through which

to raise the pressure to the desired point. It is a situation for which no immediate remedy is available.

During the next twenty years a considerable amount of progress was made in the solution of these problems, but the experimental methods that were available required elaborate and expensive equipment, and consequently the work did not advance so rapidly as might have been expected.

Satisfying progress in geophysics and geochemistry depends to a great extent on the widening of facilities for reproducing those extreme conditions which nature has utilized in producing the igneous and metamorphic rocks. The extension of these facilities has occupied a large proportion of the attention of the Geophysical Laboratory, and many of the contributions of the Laboratory have resulted directly from such efforts. Results on the systems granite -water and magnesia-alumina-silicawater, to be described farther on, can be attributed to such a widening of facilities. The equipment which made this study feasible was first described in Year Book No. 46, five years ago. Since that time similar apparatus has been installed by several universities and government laboratories here and abroad, and is now manufactured commercially by one scientific instrument company.

If a survey were to be conducted among geologists as to the most important and petrologically most interesting three-component system, it seems certain that the system quartz—soda feldspar—potash feldspar would receive by far the greatest number of ayes, for this system embraces compositions closely approaching those of the granites, the most studied and genetically the most controversial of the igneous rocks. One may fairly inquire why, if this system is of such importance, it was not investigated long ago along with the numerous

ternary systems studied in great detail since the founding of the Laboratory some forty-five years ago. The rejoinder is that this combination of oxides has such an enormous viscosity that it has heretofore resisted all attempts to decipher its equilibrium relations in the most important portion of the system. In this region, lying in general below 1000° C, the liquids are so viscous that crystallization in the dry way for periods exceeding ten years has failed to produce evidence of crystal growth from glasses of the appropriate compositions. Now, it has long been known that the viscosity of silicate melts could be lowered by the addition of volatile materials such as water, thereby greatly increasing the rate of crystallization. In the case of granitic liquids, however, relatively high pressures are required to promote solution of the low-melting substances. This high pressure coupled with the high temperature required to melt granite has been the principal deterrent to progress on equilibrium studies of these compositions. The apparatus mentioned above has cleared away these obstacles, as pressures up to 1000 atmospheres can be utilized at temperatures up to 1050° C and pressures of 4000 atmospheres have been used at 800° C. Moreover, the equipment is of simple design, and quenching experiments can be carried out with little more effort or expense than that required for the "dry" quenching studies which have been so successfully utilized by the Laboratory during the past forty years.

It would be misleading to assert that the new apparatus alone has been responsible for the rapid progress in study of these viscous mixtures. The foundation for these studies has been under construction throughout the entire existence of the Laboratory; the first publication of the Laboratory (1905), entitled *The Isomorphism and*

Thermal Properties of the Feldspars, may well be termed the footing to which subsequent papers, too numerous to report here, have added brick by brick.

Returning to the granite-water studies: it can be reported that equilibrium relations in the quaternary system NaAlSi₃O₈ -KAlSi₃O₈-SiO₂-H₂O have been studied at pressures up to 4000 atmospheres. These investigations include results on the bounding ternary systems NaAlSi₃O₈ $-KAlSi_3O_8-H_2O_5$ NaAlSi₃O₈—SiO₂ -H₂O, and KAlSi₃O₈-SiO₂-H₂O. Perhaps the most important feature of the equilibrium relations in this system is the composition of the minimum-melting mixture, which, at low pressure of water vapor, coincides almost exactly with the average compositions of the granitic rocks. This suggests that the minimum-melting composition has played an important role in the formation of granites, for there is no other known mechanism for giving the constant proportion of the two feldspars and quartz.

Studies in the system magnesia—alumina—silica—water have been carried to the publication stage this past year. This work

was also made possible by the new equipment mentioned above, and is of great significance in our understanding of the metamorphic rocks and the processes whereby they are produced from argillaceous sediments, the most important group of rocks with which the metamorphic petrologist is concerned. One of the outstanding results of the studies is the evidence indicating that the mineral assemblages that develop during metamorphism of argillaceous sediments are profoundly affected by the amount of water present. It has been demonstrated that certain important metamorphic minerals such as pyrope garnet and anthophyllite are unstable in the presence of water. This leads to the concept of "water deficient" and "excess water" assemblages. By means of the experimental work on the "excess water" region it has been demonstrated that the accepted mineral assemblages which are believed to be critical in defining the various metamorphic facies may form under identical pressure-temperature conditions.

A more detailed account of the past year's work follows.

SILICATES IN THE PRESENCE OF WATER UNDER PRESSURE

The system NaAlSi₃O₈—KAlSi₃O₈—SiO₂ (synthetic granites) (Tuttle and Bowen). In Year Book No. 50 (1950–1951) the earlier results of studies in this system were reported. Investigations have now been extended to higher pressures of water vapor, namely 4000 atm; additional results have been obtained at lower pressures where certain matters required clarification; and some of the measurements at lower pressures have been refined. The whole investigation is now about ready to be put into form for extended publication. Since the principal minerals of granites, especially the alkali granites, are the two

alkali feldspars, albite (NaAlSi₃O₈) and orthoclase (KAlSi₃O₈), together with quartz (SiO₂), the synthetic mixtures investigated may appropriately be referred to as synthetic granites.

Our earlier studies have shown that the two alkali feldspars form a complete series of solid solutions of the type with a minimum melting or crystallization temperature, but at lower temperatures these solid solutions unmix so that there is a definite unmixing curve or *solvus* giving the composition of the two feldspars in equilibrium at any temperature, the one feldspar rich in sodium, the other in potassium.

The maximum temperature on the solvus is 660°.

The lowering of the melting temperatures (crystallization temperatures) of these feldspar solid solutions under high pressure of water vapor has been reported, and is depicted in figure 2 on page 39, Year Book No. 49 (1949–1950). This figure includes the solvus also, and shows that it is unchanged by high pressure of water vapor, for the reason that the water does not play any part in the equilibrium between the solid feldspars.

If the lowering of the melting or crystallization temperature of the feldspars were carried further, eventually temperatures as low as or lower than 660° would be attained, and then two feldspars would crystallize side by side, as primary crystals from the liquid. As we have formerly reported, we have not succeeded in applying a sufficiently high pressure of water vapor to lower crystallization to such temperatures; and now that we have extended the pressure range to 4000 atm, the situation remains unchanged in that respect. At 4000 atm the feldspar minimum is at 730°, still some 70° above the maximum on the solvus.

With the addition of quartz (SiO₂) to the feldspars there is naturally a lowering of melting (crystallization) temperatures, and it was at first expected that we should quickly get down to temperatures where two feldspars would crystallize, but this expectation has not been realized. In figure I is shown the liquidus diagram for 1000 atm pressure of H2O, and the temperature contours show that the minimummelting quartz-feldspar composition melts at 720°. Even a pressure of 3000 atm is not quite sufficient to lower the minimum temperature to 660°. The lowering effect of increased pressure of water vapor is a rapidly diminishing quantity, but increase of pressure to 4000 atm does bring the

temperature barely below the 660° value (655°), and in accordance with theoretical expectation two feldspars do separate side by side from the liquid. Since the top of the solvus is flat, the two feldspars are already notably different in composition even at 655°. With further lowering of crystallization temperature, consequent upon further increase of water pressure, it can be predicted that the two feldspars will diverge in composition in accordance with the solvus relations; but it requires a very large increase in pressure to produce significant additional lowering. The 4000 atm pressure corresponds to the superincumbent load at a depth of some 9 miles. Moreover, pressure as such is not enough; there must be an adequate supply of water so that the water pressure has the value indicated, and the liquid at this pressure is capable of dissolving some 9 per cent water. If only a smaller quantity of water is available, the full lowering effect on the melting temperature will not be realized even though the applied load is the full 4000 atm.

It is improbable that many granites reaching the light of day have crystallized at depths greater than 9 miles, and if a few have, it must be regarded as very unlikely that the liquid magma contained the oper cent or more of water necessary to bring the crystallization temperature down to a value at which two not very different alkali feldspars would form as primary crystals from the liquid. To bring the temperature of crystallization down to a value at which nearly pure albite and comparatively pure orthoclase would form directly from the liquid requires conditions of pressure and water content that are beyond all reasonable assumption.

The experimental findings thus forcibly raise the question whether two feldspars did originally crystallize from most granite magmas or whether they were initially in

mutual solid solution as a single feldspar which became two feldspars only through changes subsequent to the complete or almost complete crystallization of the granite. evidence is found pointing to the fact that in a great many instances there was initially but one feldspar and that the separation into two has been a progressive process passing through the stages of cryp-

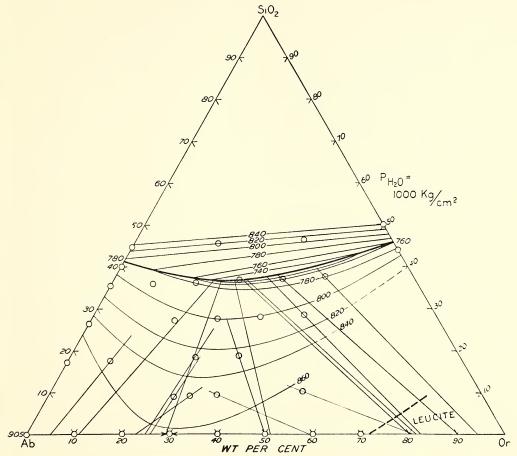


Fig. 1. Crystallization diagram, under 1000 atm pressure of H_2O , of mixtures of albite, orthoclase, and quartz, with temperature contours of the liquidus (crystallization) surface which determine its minimum temperature (720°) and the position of the minimum-melting composition.

It is possible to shed much light on this subject by examining the feldspars of the quickly chilled volcanic equivalents of granites, namely the rhyolites, and also by comparing the feldspars of small granite masses with those of larger masses, and especially the feldspars of border facies of large intrusives with those of the interior of such masses. When this is done, much

toperthite, microperthite, perthite, and finally a granular fabric. The stage attained depends on the opportunity for unmixing on cooling, largely a question of time available, but presumably also affected by movements inducing strain and by degree of retention of pore-space water vapor to serve as a recrystallizing medium. The whole situation is complicated by the

fact that in the natural rock there is always at least a little lime feldspar to enter into solid solution, and its presence raises the temperature at which the two feldspars may form. This relation has been ascertained experimentally in a qualitative way, but the exact quantitative effects remain to be investigated. It is therefore to be expected—and a study of rocks bears out that expectation—that as the proportion of lime feldspar increases in the granite-granodiorite clan, the likelihood increases that a rock will have two feldspars initially crystallized as such. Our studies, both of the synthetic mixtures and of natural rocks, are now directed toward elucidation of these problems, with the quantitative element in mind.

Composition of natural granites and its relation to the minimum-melting synthetic mixture. It was demonstrated some years ago that the average composition of granites corresponds very closely to the composition of the minimum-melting mixture in the system NaAlSi₃O₈—KAlSi₃O₈— SiO₂ as then determined. This fact has been advanced as a strong argument in favor of the thesis that granites are the product of crystallization of the low-melting residuum of fractional crystallization of magmas, or of initial re-fusion of granitelike material. The fact has, moreover, been advanced as an argument against replacement as the major prevalent factor in the origin of granites, inasmuch as replacement, unlike fractional crystallization, offers no rationale for the tendency toward the balanced proportion of the constituent minerals which the rocks actually show.

It is now possible to examine these arguments more closely. The recent experimentation provides reliable information as to the composition of the minimum-melting liquid and its variation in the presence of varying water content. Especially in the case of granite, such information must be

regarded as pertaining directly to the problem in hand, for it is plain enough that granite magmas always contain some water.

In addition, we have made a new approach to the question of the average composition of granite, or, more specifically, of the average proportions of the components of lowest melting composition. The average used in the earlier attack was simply the arithmetic mean of the normative components-albite, orthoclase, and quartz-of all rocks showing more than 80 per cent of these components. No information was thus gained as to the extent of departure of individual granites or of their average departure from the mean, to which great significance has been attached. In the new method the proportions of the normative constituents are plotted on triangular co-ordinates for each individual rock meeting the above requirement. Each rock is thus represented by a point, and the diagram is then contoured in a manner which indicates the number of points in equal areas. Such a diagram offers a graphic picture of the position of greatest concentration of points and of the extent to which granite compositions are concentrated near this position. Figure 2 is the actual diagram so constructed.

Even taken by itself, figure 2 illustrates the enormous extent to which granite formation must be dominated by factors tending to give the rock a nearly constant composition, and demonstrates the minor role played by processes of replacement which would give random relative proportions of the constituents. Moreover, when the actual composition range in which granites are most highly concentrated is compared with the minimum-melting compositions as determined experimentally, the degree of correspondence is so great as to fortify strongly the original conclusion based on the preliminary results obtained with dry-

melt studies and on the mere arithmetic mean of granite compositions.

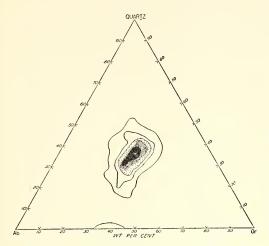


FIG. 2. Contoured frequency-distribution diagram of the normative composition (albite + orthoclase + quartz) of 571 analyzed rocks (granites and syenites) containing 80 per cent or more of these normative constituents. 0.25 per cent counter; contours > 1, > 2, > 3, > 4, > (5-7) per cent.

The system $MgO-Al_2O_3-SiO_2-H_2O$ (Yoder). The largest group of rocks with which the metamorphic petrologist must deal is that developed during the metamorphism of the argillaceous sediments. The composition of the bulk of these sediments can be closely approached by mixtures in the magnesia—alumina—silica—water system. Therefore, a study of that system is pertinent to the understanding of the origin of many metamorphic rocks.

Some of the important minerals which occur in the system are talc, pyrophyllite, serpentine, clinochlore, cordierite, forsterite, enstatite, spinel, brucite, periclase, quartz, and anthophyllite. A new low-temperature form, aluminous-serpentine, having the clinochlore composition, was prepared, but its stability has not been determined with certainty. It is suspected that this form may be isostructural with one form of serpentine. Pyrope was not

synthesized, and natural pyrope was not found to be stable in the presence of an excess of water vapor in the pressure and temperature range investigated. Amesite and magnesian chamosite were not produced. The existence of two modifications of cordierite (Rankin and Merwin, 1918) was confirmed. Metastable growth of many of the phases occurring in the system is common, and these phases survive for long periods of time.

Equilibria in a portion of the system were determined at temperatures from 430° to 990° C at pressures up to 2000 atm. The univariant pressure-temperature curves were bracketed for the following reactions:

and some of the previously investigated reactions in the MgO-SiO₂-H₂O system (Bowen, Tuttle) are confirmed. All the pressure-temperature curves are given in figure 3. The phase assemblages obtained in equilibrium with vapor are summarized at significant temperature intervals at 1000 atm (15,000 pounds per square inch) in figure 4. These assemblages outline compositions in the tetrahedron for which water is prohibited as a stable phase. For this reason the "water-deficient region" and the "excess-water region" are defined. It is shown that some minerals—for example, pyrope and anthophyllite-may be stable only in the water-deficient region. It is concluded, therefore, that the presence of an "excess" or "deficiency" of water vapor greatly influences the mineralogy of a metamorphic rock. On the basis of the assemblages obtained experimentally in the presence of an excess of water, and of the concept of the water-deficient region, it is

demonstrated that all the now-accepted critical mineral assemblages which have been used to define the various metamorphic facies may have formed under the same pressure and temperature conditions, the different facies being primarily

that the reactions are essentially insensitive to pressure. This relation can probably be regarded as extending to the highest pressures in the earth's crust, approximately 10,000 atm or 150,000 psi. It is believed, therefore, that pressure does not

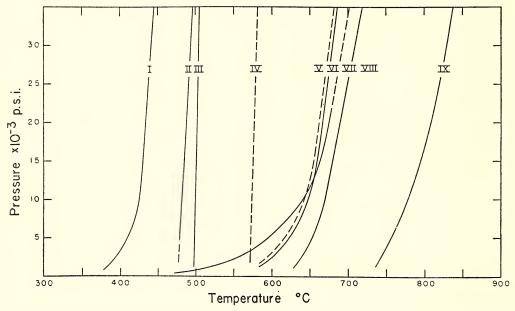


Fig. 3. Some of the pressure-temperature curves of univariant equilibrium in the system MgO $-Al_2O_3$ — SiO_2 — H_2O . I, $S+B \rightleftharpoons F+V$; II, $B+Al-S \rightleftharpoons F+Sp+V$; III, $S \rightleftharpoons F+T+V$; IV, Py $\rightleftarrows Mu+Cr+V$; V, $Cl+T \rightleftharpoons F+Co+V$; VI, $F+T \rightleftharpoons E+V$; VII, $B \rightleftharpoons P+V$; VIII, $Cl \leftrightharpoons F+Co+Sp+V$; IX, $T \rightleftharpoons E+Q+V$. The assemblages stable in the divariant regions between curves are given in figure 4.

Abbreviations for figures 3 and 4: A, andalusite; Al-S, aluminous serpentine; B, brucite; C, corundum; Cl, clinochlore; Co, cordierite; Cr, cristobalite; D, diaspore; E, enstatite; F, forsterite; K, kyanite; Ka, kaolinite; M, montmorillonite; Mu, mullite; P, periclase; Py, pyrophyllite; Q, quartz; S, serpentine; Sil, sillimanite; Sp, spinel; T, talc; Tr, tridymite; V, vapor.

a function of the bulk composition. A new set of critical assemblages indicative of significant pressure and temperature conditions is required, therefore, for the various stages in progressive metamorphism. The selection of these critical assemblages will most likely have to await further experimental work.

Another conclusion may be mentioned in connection with the shape of the pressure-temperature curves given in figure 3. A cursory glance at the curves indicates

play a significant role in the MgO—Al₂O₃ —SiO₂—H₂O system, except in the very lowest pressure regions. Previous belief held that the greatest changes in the mineralogy of metamorphic rocks were to be expected at the highest pressures.

The observation that the minerals in the MgO—Al₂O₃—SiO₂—H₂O system grow at different rates led to a reconsideration of the concept of the isograd as defined by Tilley (1924). It was concluded that there are two different types of isograd. One

represents the attainment of a univariant condition and in this sense conforms to the original definition. The other is dependent on the attainment of an adequate rate of growth. (It is suggested that the latter be called *isoblast*.) The present sillimanite isograd is of the first type. The chlorite, garnet, and biotite "isograds" are

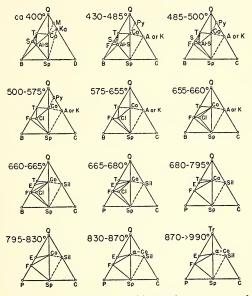


Fig. 4. Assemblages stable in the presence of an excess of water vapor at significant temperature intervals and 15,000 psi in the system MgO—Al₂O₃—SiO₂—H₂O. For abbreviations see legend to figure 3.

probably of the second type. It is recommended, therefore, that only polymorphic transitions and mineral decompositions be used as indicators of grade of metamorphism. The appearance of a mineral, although useful locally, cannot be used with assurance as a measure of the attainment of specific pressure and temperature conditions in widely separated areas.

This is the first investigation of a system which can be applied directly to a large group of common metamorphic rocks.

The system H_2O — Na_2O — SiO_2 (Morey, Hesselgesser). The complete solubility

curve of a salt in water covers the entire range in composition and temperature from the melting point of ice to the melting point of the salt. When the melting point of the salt is high, the vapor pressure of the saturated solutions may reach high values, and the solubility of the solid in the gas may become so great as to be not only of theoretical interest, but also of practical importance. There is almost no experimental knowledge of systems of this kind that interest the geologist, and this study is an introduction to an almost new field, one important to geochemistry. Knowledge concerning systems containing water at high temperature and pressure is essential to an understanding of the mechanism of rock formation, and information about the gas compositions which may coexist with aqueous silicate melts under high pressure is essential to understanding of the genesis of vein minerals, pegmatites, and pneumatolytic processes in general.

A method and an apparatus have been devised which have made possible the analyses of coexisting gas and liquid phases in the system H₂O-Na₂O-SiO₂. The isothermal polybaric saturation curves -both gaseous and liquid-of quartz, sodium disilicate, and sodium metasilicate have been determined at 400° C, and the melting pressure of Na₂O·2SiO₂ was found to be 2200 atm. The boundary curve gas + liquid + quartz + sodium disilicate is continuous and does not intersect the critical end-point curve, and it is likely that the boundary curve gas + liquid + sodium disilicate + sodium metasilicate has a similar relationship. There are probably two critical regions surrounded by critical curves, one of which is represented by (gas = liquid) + Na₂O·SiO₂, the other by (gas + liquid) + Na₂O·2SiO₂, although the possibility has not been excluded that these two critical curves intersect, giving rise to a double critical end point (gas = liquid) + Na₂O·SiO₂ + Na₂O·2SiO₂.

Solubility in superheated steam at high pressures (Morey). A new method for measuring the solubility of solid materials in steam at high pressures and temperatures was described in Year Book No. 48 (1948–1949). Since that time the solubility of numerous silicates and other inorganic compounds has been determined. This past year the solubility of CaCO₃ in steam has

been measured, and at 500° C and 1000 atm was found to be 15×10^{-6} , or 0.0015 per cent. A similar solubility has been found for CaSO₄. Experiments with CaCO₃ are also being made with H₂O saturated with CO₂ under the pressure of liquid CO₂ at room temperature. These results have a direct bearing on the vapor transport of solids, a phenomenon which has long been considered by field geologists to be of major importance in the formation of many ore deposits.

X-RAY INVESTIGATIONS

The use of X-ray methods continues to provide an important tool for many of our researches on silicates. Such methods have proved indispensable in the study of the feldspars, a group of minerals that is receiving renewed attention in our postwar program. We have been limited to powder methods in recent years, but the situation has been rectified in the past year, since Dr. J. V. Smith joined the staff. The laboratory for single-crystal studies has now been placed in running order again and some new equipment added. Facilities have been set up for calculation of Fourier series.

Single-crystal studies on paracelsian (BaAl₂Si₂O₈) (Smith). Last year we reported on two new synthetic modifications of the feldspar anorthite (CaAl₂Si₂O₈). One type is orthorhombic and the other hexagonal. Both forms are believed to be unstable and, presumably for this reason, are not found occurring naturally. The hexagonal modification may be isostructural with α-celsian, a hexagonal form of barium feldspar. The possibility that the orthorhombic form might be isostructural with paracelsian, another modification of the barium feldspar, has been considered, but as there were no X-ray data on paracelsian, single crystals were obtained and studied. Crystals of paracelsian were examined by single-crystal and powder X-ray methods, with the following results:

From precession photographs (not assumed to be orthorhombic): $a \sin \beta = 9.02 \pm 0.09 \text{ Å}$ $b = 9.50 \pm 0.09 \text{ Å}$ $c \sin \beta = 8.47 \pm 0.09 \text{ Å}$ $\beta = 90^{\circ} \pm 0.5^{\circ}$

From Geiger-counter spectrometer (assumed to be orthorhombic): $a^1 = 9.076 \pm 0.005 \text{ Å}$ $b = 9.583 \pm 0.005 \text{ Å}$ $c^1 = 8.578 \pm 0.005 \text{ Å}$

There are 4 units of BaO·Al₂O₃·2SiO₂ in the unit cell. Paracelsian is monoclinic but very markedly pseudo-orthorhombic, with β very near to 90°. These observations confirm the goniometric and optical measurements of L. J. Spencer, who found that paracelsian was optically monoclinic but morphologically orthorhombic. atic intensity absences occurred only for reflections with h odd in (hoo), k odd in (oko), I odd in (ool), h odd in (hol). In addition, (k+1) odd in (okl) is a very pronounced pseudo absence. The space group is therefore P21/a, assuming that reflections with k odd in (oko) are truly absent; if not, the space group is P2/a or Pa. The crystals have the pseudo space group Pna or Pnam.

Spencer pointed out the "remarkable agreement between the angles of paracelsian and those of topaz and danburite." Comparison of the X-ray data shows that there can be no structural relation between topaz and paracelsian. There exists, however, a close relation between paracelsian, BaAl₂Si₂O₈, and danburite, CaB₂Si₂O₈. The structure of danburite is known, and a similar structure for paracelsian was proposed, with Ba substituting for Ca and (Al+Si) for (B+Si).

A complete set of intensity data for the [100] [010] [001] zones of paracelsian was collected by accurate single-crystal, eye-estimation techniques. The resulting structure amplitudes were compared with those calculated for the proposed structure. There was no agreement. After the atomic co-ordinates were changed by a few hundredths, however, good agreement was obtained. Refinement of parameters using F_o and $(F_o - F_c)$ syntheses is in progress.

The structure of melilite (Smith). A high-accuracy structure determination has been carried out on this interesting solidsolution series to provide a basis for crystal chemistry arguments concerning substitution of certain atoms in melilite. The structure provides some much needed accurate data on the calcium polyhedron. The calcium atom has four near and four distant oxygen atoms. The polyhedron is much distorted and very different from the normal polyhedron. Also, there is a large deviation from Pauling's rule of electrostatic valence bonds. Further studies will be attempted to elucidate this unusually large deviation from Pauling's rules.

Distortions of the O-O distances are probably the result of the sharing of edges between polyhedra, but the distortion in the Si-O distance cannot be explained by the usual theories. Perhaps it is related to the deviation from Pauling's rules, and a large fraction of each bond is covalent rather than ionic.

Study of composition of natural and synthetic olivine by X-ray methods (Yoder). A series of natural and synthetic olivines has been studied by the powder method. It was found that composition can be estimated to ±2 mol per cent in terms of the major end members of the group (forsterite, fayalite, and tephroite) by measuring the spacing of planes particularly sensitive to changes in composition. Olivines in common igneous rocks are variable in composition, and the variation usually changes systematically with regard to the nature of the igneous body. This new method of obtaining the composition of the olivines will greatly aid the petrologist in studying those olivine-bearing igneous bodies for which a large number of tedious and expensive chemical analyses are usually required.

Orientation of crystals on the precession camera (Smith). Two methods for orienting crystals on a precession camera have been developed. One method utilizes an auxiliary telescope which is built onto the camera and permits using morphological properties in aligning crystals; the other method makes use of a Laue photograph instead of the usual small-angle precession photograph. Under some conditions the Laue photograph is easier to interpret.

X-ray study of deformed quartz crystals (Smith, Tuttle). Quartz grains in deformed rocks are known from optical studies to show a remarkable preferred orientation. The pattern of the orientation of the optic axes (c-axis) may be represented by a single grouping of axes along an axis of the deforming stress pattern, or the orientation may be highly complex and show an astonishing variety of patterns.

Numerous theories have been proposed to explain the quartz orientation on the basis of twinning, translation, and fracture, but no completely satisfactory hypothesis has evolved.

Throughout the Washington, D. C. area the ubiquitous mica schists have suffered a deformation which was arrested in an early stage and is now represented by small folds containing quartz which has been only partially reoriented by the folding. These small drag folds offer an exceptional opportunity for investigating the problem of quartz fabrics. Accordingly, single-crystal X-ray studies are now under way on oriented quartz crystals from these folds.

Preliminary results on three crystals indicate that each consists of many crystal units all of which have approximately the same orientation. The range of orientation of these units has been measured by a new method that involves rotation of the crystal together with an identical rotation of the photographic film. On the resulting film the range of crystal orientation (after linear corrections) is shown as it would appear if plotted on a stereographic projection. Tentative results are: (1) The crystals appear to consist of many units, all having approximately the same orientation; the range of orientation of different crystal face normals is from 5° to 30°. (2) No evidence of twinning was discovered. (3) The orientation spread of the crystals is not isotropic; the direction of maximum spread lies approximately in a plane perpendicular to the fold axis.

Further work is in progress, and it is anticipated that new light will be thrown on the foremost problem of structural petrology: the mechanism producing quartz orientation in tectonites.

High-temperature alkali feldspars (Donnay). The cell dimensions of the triclinic compositions, from Ab₆₅Or₃₅ to Ab₁₀₀ in

the albite-orthoclase series, have been determined from powder patterns taken on the Philips back-reflection X-ray spectrometer (Adams). Cell dimensions obtained last year for the monoclinic compositions (Or₁₀₀ to Ab₆₅Or₈₅) have been refined. The significance of this change in symmetry near Ab₆₅Or₈₅ is discussed in the following section (MacKenzie). Theoretically, there should be a range of compositions (at constant temperature) in which the monoclinic and triclinic forms coexist; no such two-phase region was discovered.

The superstructure in plagioclases (Donnay). Superstructure lines on c-axis rotation photographs and on precession photographs have been reported in the literature for compositions ranging from $Ab_{40}An_{60}$ to An_{100} .

The effect of the thermal history of the crystal on the appearance of these superstructure lines has been investigated on four natural crystals having a composition of approximately Ab₃₅An₆₅. The following results were obtained: High-temperature crystals show no superstructure lines after exposure times greatly exceeding those mentioned in the literature, whereas low-temperature crystals show the superstructure lines after 8-hour exposures. These results can be explained on the basis of ordering: superstructure lines indicate a large true cell, multiple of a small pseudo cell; low temperature indicates slower rate of crystallization and more opportunity for ordering; at high temperature, disorder prevails and the small pseudo cell becomes the true cellhence, no superstructure lines.

More experimental work is needed. It is hoped that the temperature of formation can be correlated with the degree of ordering. Such correlation, if confirmed, should be looked for in other minerals. Ordering may well be of practical importance as a geologic thermometer.

The crystal structure of hexagonal CaAl₂Si₂O₈ (Donnay). The possibility that hexagonal CaAl₂Si₂O₈ might be isostructural with α-celsian, BaAl₂Si₂O₈, has been investigated. Structure-factor calculations have ruled out this possibility. However, α-celsian has a high-low inversion which is absent in the lime compound, and the possibility still exists that hexagonal CaAl₂Si₂O₈ is isostructural with the high-temperature modification of α-celsian.

Preliminary results are awaiting publication. The crystal symmetry obtained from X-ray photographs is too high to be compatible with a reasonable structure; it is presumably to be accounted for by merohedral twinning, which could escape X-ray detection. Long exposures (36 hours) have now revealed disorder: the reciprocallattice modes *lll* are drawn out into a streak, and the *c* unit length of 7.358 Å is a pseudo period, which must be multiplied by 2.

Soluble silicates (Donnay). Cell dimensions of the high-temperature modification of sodium disilicate have been refined from a powder chart: $a=15.45\pm0.03$, $b=4.909\pm0.005$, $c=6.428\pm0.015$ Å. The calculated and observed densities are 2.48 and 2.47 g/cm³, respectively.

Two lithium silicates prepared by Kracek have been studied by single-crystal methods (Weissenberg and precession) with the following results: Low-temperature Li₂Si₂O₅ is orthorhombic, with space group Ccc2 and 4 formula units per cell, a=5.80, b=14.66, c=4.806 Å, all ± 0.3 per cent. Density 2.438 (calc.), 2.454 (obs.). The crystals are platy on (010). This compound has been reported as monoclinic. Li₂SiO₃ is orthorhombic, with possible space group Ccmm, Ccm2, or Cc2m. There are 4 formula units in the cell: a= 5.43 ± 0.02 , $b = 9.41 \pm 0.03$, $c = 4.660 \pm 0.015$ Å. Density 2.50 (calc.), 2.520 (obs.). The crystals are roughly lath-shaped, tabular on (010), elongated along c. This compound has been reported as being hexagonal.

FELDSPARS AND GRANITE

Significance of the differences between modal and normative feldspar of granites (Tuttle). As was mentioned above, phaseequilibrium studies on the alkali feldspars, albite and orthoclase, have demonstrated that at high temperatures these two feldspars form a complete series of solid solutions of the type with a minimum melting or crystallization temperature, but that at lower temperatures the solid solution becomes unstable and unmixing into two phases takes place. Evidence for this unmixing process in natural rocks is found in the perthosites (rocks consisting almost entirely of an alkali feldspar, perthite, which usually occurs as a fine lamellar intergrowth of albite and orthoclase) and the perthite-quartz granites. These perthite-

bearing rocks offer convincing evidence for the former presence of a single homogeneous feldspar. On the other hand, many granites having essentially the same bulk composition as the perthite-quartz granites consist of albite, orthoclase, and quartz, with no textural evidence that the two feldspars had unmixed from a homogeneous phase. It has been suggested that the latter type of granite may have crystallized as a one-feldspar granite (perthite + quartz), but that subsequent unmixing has more or less completely separated the two feldspars by a process of solid diffusion and recrystallization, probably aided by volatile materials concentrated during crystallization. Evidence for this unmixing episode might be discovered at the more

quickly cooled contacts of the two feldspar granites.

Accordingly, specimens from granite contacts have been collected in Scotland and Norway; and modal analyses, using the methods developed by Chayes (Year Book No. 47, 1947–1948), have been made on about fifty specimens. These modes are shown in figure 5, which has

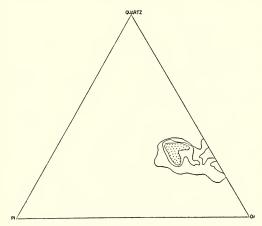


Fig. 5. Contoured frequency distribution diagram showing modal composition of granites from Scotland and Norway. 49 analyses; 0.25 per cent counter; contours >0, >4, >8 per cent.

been prepared in the same manner as figure 2. A comparison of this diagram with figure 2 in the section on the modal composition of granites in last year's report reveals remarkable differences in the amounts of the various mineral species present. Chemical analyses of representative samples of these specimens, which are mostly contact samples, indicate that chemically these granites are essentially identical with those studied by Chayes. These results show that although a granite may consist, chemically (or normatively), of approximately equal amounts of albite, orthoclase, and quartz, mineralogically there may be all gradations from one feldspar and quartz, all the albite being in solid solution in the orthoclase, to two feldspars and quartz, the albite phase occurring as separate, discrete crystals. This finding supports the suggestion that unmixing may be a dominant process in the evolution of the granitic texture.

Composition of rock-forming microcline (Chayes). Our knowledge of the chemical composition of nonvolcanic potash feldspar and acid plagioclase is based almost entirely on specimens from pegmatites and veins. Compositional information concerning these feldspars is lacking for the granitic rocks. Accordingly, we have undertaken to separate the alkali feldspar from a suite of granites with known modal composition.

The separation involves crushing about 40 grams of rock, the 100–150 mesh fraction of which usually yields 1 to 2 grams of high-purity alkali feldspar. Separation is carried out by heavy liquids, and the purity of the concentrate is checked by X-ray and optical examination. The final concentrate will be analyzed for alkalis by the flame photometer, and partial chemical analysis of a few samples will be made by other methods. It is anticipated that much useful mineralogical information will be generated as a by-product of this program.

Modal analysis of granite (Chayes). The modal analysis of granitic rocks is proving to be a powerful tool for our renewed attack on problems concerning the origin of granite. Results on 260 specimens of granite from New England, Texas, and the Southeast reveal a remarkable concentration, as shown on a contoured frequency diagram (Year Book No. 50, 1950-1951), about a composition having approximately a 1:1:1 ratio of plagioclase: orthoclase: quartz. The proximity of this modal maximum to the minimum-melting composition in the system albite—orthoclase quartz, as determined by Bowen and Tuttle (fig. 1), is certainly more than a coincidence, and it offers further evidence of the control of granite composition by the minimum-melting composition.

Optical and X-ray studies of alkali feldspars (MacKenzie). Alkali feldspar investigations are continuing, with the objective of elucidating the stability relations of the various modifications and determining chemical composition by X-ray and optical methods.

In the high-temperature alkali feldspar series (sanidine—anorthoclase—high bite), the optical investigation consisted of accurate measurements of the optic axial angle in at least six individual crystals or cleavage fragments of each mineral sample, both in the natural state and after heating at 900° C for approximately 24 hours. In the case of the triclinic members of the series, the orientation of the optic plane was also determined. The effect of prolonged heating on the size of the optic angle and on the orientation of the optic plane is being investigated. The bulk composition of each sample—and, in the case of unmixed samples, the composition of the individual phases—has been determined, using the Philips X-ray spectrometer by the method described by Bowen and Tuttle (1950). All together, 30 natural high-temperature alkali feldspars have been investigated in this manner; of these, 23 have been analyzed chemically, and it is hoped to analyze the remainder during the coming year.

G. Donnay has shown that, by inspection of the X-ray spectrometer pattern of a high-temperature alkali feldspar, the symmetry can easily be determined from the fact that in the case of a triclinic mineral two peaks representing reflections from the (111) and (111) can be recognized, whereas only one peak, representing both (111) and (111), is present in the case of a monoclinic mineral. The most sensitive optical criterion of triclinic symmetry is

the presence of multiple twinning, which is generally according to both the albite and pericline laws. The change of symmetry at about Or35Ab65 has been difficult to explain, since there is no apparent discontinuity in the liquidus curve of the series, and a solid-solution series involving a change in symmetry is unknown in other silicate systems. Bowen and Tuttle (1950) were of the opinion that in the case of high albite there exists a nonquenchable transformation which, with increasing temperature, would result in the gradual change of cell constants to monoclinic symmetry. The triclinic members of the high-temperature alkali feldspar series were investigated by both X-ray and optical means at elevated temperatures, and the results obtained confirm the belief that a gradual change in cell constants takes place, resulting in a cell with monoclinic symmetry. The X-ray investigation was carried out by comparison of X-ray powder diffraction patterns recorded at elevated temperatures, using a small furnace suitable for mounting on the Philips backreflection X-ray spectrometer. For the optical investigation, the disappearance on heating and the reappearance on cooling of the twinning lamellae was used as the criterion of the change in symmetry. Bowen and Tuttle had previously detected very slight difference in the X-ray diffraction patterns of the soda-rich plagioclases according to the temperatures at which they had been crystallized. Confirmation of these differences was obtained as a result of the present investigation, in which it was found that synthetic sodic feldspars crystallized "dry" differed, as regards the temperature at which they became monoclinic, from those crystallized at lower temperatures hydrothermally. Heating the hydrothermally crystallized material for 7 days at 1080° C made it identical with the "dry" material.

No detailed optical work has yet been done on the high-sanidine-high-albite series, but one mineral sample has been found in which crystals are sometimes formed to high sanidine by prolonged heating has been known for a considerable time, but the relations between microcline, orthoclase, and adularia are still completely



Fig. 6. Powder diffraction charts of six low-temperature alkali feldspars (compositions given as weight per cent). Dashed lines illustrate the change of the 130 and 130, 131 and 131 spacings in the triclinic feldspars. Curve A, Or_{84.5}Ab_{13.8}An_{1.7}, orthoclase; B, Or_{81.5}Ab_{18.1}An_{0.4}, orthoclase; C, Or_{85.9}Ab_{12.7}An_{1.4}, microcline; D, Or_{83.8}Ab_{14.9}An_{1.8}, microcline; E, Or_{90.5}Ab_{9.3}An_{0.2}, microcline; F, Or_{88.5}Ab_{10.6}An_{0.9}, orthoclase and microcline.

zoned from sanidine to high sanidine. This type of zoning must be extremely rare, for it is apparently not recorded in the literature. The fact that alkali feldspars at the potash end of the series can be trans-

unknown, and all attempts to transform one form into another have failed.

It has been found that orthoclase can very easily be distinguished from microcline by X-ray powder diffraction patterns, and, in two samples of analyzed material, peaks characteristic of both orthoclase and microcline have been identified. G. Donnay has shown that for the triclinic high-temperature alkali feldspars there is a continuous change in crystallographic constants, with substitution of Na for K. This does not, however, appear to be the case for the microclines. Figure 6 shows the X-ray powder patterns of a series of six

low-temperature alkali feldspars arranged to show the change in the separation of the 130 and 130 peaks and the 131 and 131 peaks. From these charts it would appear that there is no simple relation between the chemical composition and the crystallographic constants. Determination of accurate cell constants from these charts is in progress and may throw more light on this question.

ANHYDROUS SILICATE SYSTEMS

The quaternary system K₂O-MgO-Al₂O₃—SiO₂ (Schairer, Thwaite). All compositions in the volume leucite-corundum-spinel-silica, during crystallization, proceed toward a quaternary eutectic where the crystalline phases are potash feldspar, silica, mullite, and cordierite, and those in the volume leucite-forsterite-spinelsilica proceed toward a quaternary eutectic where the crystalline phases are potash feldspar, silica, enstatite, and cordierite. When we examine the compositions of these two quaternary eutectics, we find they are very close to each other and to the composition of the binary eutectic between potash feldspar and silica. This binary eutectic is very close in composition to a normal potash granite. Each of the two quaternary eutectics has a rather low total magnesia content, and consequently only small amounts of cordierite and mullite or cordierite and enstatite are present in the wholly crystalline product. In natural rock-forming magmas, where some water is always present during the late stages of crystallization, we should expect to find a small amount of muscovite along with a small amount of either cordierite or biotite instead of mullite and cordierite, or small amounts of biotite and enstatite or biotite and hornblende instead of cordierite and enstatite, and to find quartz instead of tridymite because of the lower temperatures that prevail in the hydrous system.

Some of the liquids in the two volumes we have been discussing are not too far removed in composition from possible magmas. Our simplified "magmas" in K₂O-MgO-Al₂O₃-SiO₂ are, of course, anhydrous instead of having at least a small water content; there is only potash present with no soda, only magnesia present with no ferrous oxide, only alumina present with no ferric oxide, and there is no lime present. In spite of these deficiencies, we can see that a large range of rock-forming compositions would give a common end product of crystallization if differentiation proceeded by fractional crystallization. We should note the significant fact that this same end product, a potash granite, would be reached even if these "simplified magmas" were contaminated with numerous small fragmental xenoliths of basic rocks such as peridotites or dunites, or if they were contaminated by assimilation, in whole or in part, of xenoliths of highly aluminous sediments.

With perfect equilibrium between crystals and liquid during crystallization, some portions of each of the volumes leucite-corundum-spinel-silica and leucite-forsterite-spinel-silica would be completely solid before the liquid could reach a quaternary eutectic composition. With adequate fractionation during crystallization, however,

all compositions in both these volumes would yield a residual liquid closely approaching, in these soda- and lime-free compositions, those of potash granites. Thus we see that our studies of these melts in K₂O—MgO—Al₂O₃—SiO₂ add further quantitative support to Bowen's arguments for the importance in petrogenesis of KAlSiO₄—NaAlSiO₄—SiO₂ as "petrogeny's residua system."

An examination of the stable assemblages of crystalline phases present in the portion of the quaternary system we have been discussing shows that certain combinations of crystalline phases are incompatible at equilibrium. Twenty-nine incompatible pairs are noted. A word of caution should be added. These incompatibilities are at and just below temperatures where a liquid phase is present, and might be changed at lower temperatures. All these observations are of interest in metamorphic petrology.

One of the objectives of the investigation of the quaternary system K₂O-MgO-Al₂O₃—SiO₂ was to provide reliable phaseequilibrium data on a series of dry melts to which water could be added later in order to study the stability relations of phlogopite, the simplest of the dark micas (biotites). A simple empirical formula for phlogopite is K₂O·6MgO·Al₂O₃·6SiO₂· 2H2O. This composition lies in the triangle leucite-forsterite-periclase, which does not lie within the volumes whose dry crystallization has just been discussed. It was necessary, however, to obtain the phase-equilibrium data and to prepare the melts, particularly in the volume leuciteforsterite-spinel-silica, before proceeding with the study of the hydrous melts and the stability relations of dark micas.

It is always fortunate when results of pure research on geological problems can be of immediate use to industry. The data on crystallization and melting in the portion of the quaternary system just discussed predict the effects of the addition of K₂O, MgO, and SiO₂ on the refractoriness of high-alumina (mullite) refractories, and the effects of K₂O, MgO, Al₂O₃, and SiO₂ on basic (periclase or forsterite) refractories. The data provide information not only on the refractoriness, or lack thereof, of a large range of compositions, but also on the chemical as distinct from the mechanical aspects of the attack of slags on a wide range of refractories. They also furnish basic information which can be applied to many technical problems in the fields of ceramics and metallurgy.

New joins now under study. The binary eutectic between leucite and spinel has been located at 1553°±5° C and at the composition leucite 88.5, spinel 11.5 per cent. The join leucite—forsterite—spinel and the join leucite—corundum—spinel are both ternary systems with ternary eutectics, and data for these joins are now nearly completed.

The composition K₂O·6MgO·Al₂O₃·6SiO₂ (phlogopite is K₂O·6MgO·Al₂O₃·6SiO₂·2H₂O) lies in the join leucite—forsterite—periclase and has a forsterite liquidus at 1628° ±5° C. The system leucite—periclase is not binary, and a field of forsterite cuts this line. No piercing points lie in the join leucite—forsterite—periclase.

Leucite — forsterite — silica — potassium disilicate. Progress in this portion of the quaternary system is very slow. The siliceous melts are difficult to prepare and take a year or more to crystallize, and then runs of long duration must be made to attain equilibrium between crystals and liquid. It will be some time before this portion is completed.

The quaternary system $Na_2O-Al_2O_3-Fe_2O_3-SiO_2$ (Schairer, Thwaite). During the past year studies of the join nepheline—sodium disilicate—acmite were completed. The three piercing points of quaternary univariant lines in this plane were located accurately as to temperature and composi-

tion. The studies show that some molecule containing Fe₂O₃ must enter the nepheline crystal lattice, and the nepheline crystals are not pure nepheline, but complex solid solutions. Studies of the join sodium disilicate—albite—acmite were also completed, and the two piercing points of quaternary univariant lines were located accurately as to temperature and composi-Na₂O·Fe₂O₃·6SiO₂ (iron albite molecule) is present in limited amount in solid solution with Na2O·Al2O3·6SiO2 in the albite crystals. Studies of the join nepheline—5Na₂O · Fe₂O₃ · 8SiO₂—acmite were also completed, and the three piercing points of the quaternary univariant lines were located accurately as to temperature and composition. The nepheline crystals are not pure Na2O·Al2O3·2SiO2, but have albite and some molecule containing Fe₂O₃ in solid solution. During the next year the data on the five joins now completed will be analyzed and new joins will be studied to push work on this quaternary system toward completion.

The systems Na₂O—FeO—SiO₂ and Na₂O—MgO—SiO₂ (Schairer, Thwaite, Yoder). The system Na₂O—FeO—SiO₂ proves to be very interesting and complex. Unfortunately the melts are more viscous than was expected, and it is impossible to get equilibrium between crystals and liquid in an hour or less as was anticipated. At least four unknown crystalline phases have been encountered already. We have their X-ray patterns but do not yet know their chemical composition. Work on this system is still in the reconnaissance stage.

The crystalline phases in Na₂O—FeO—SiO₂ should be analogous to those in Na₂O—MgO—SiO₂. We have prepared several different compositions in the latter system that might be compounds, and they are now under study. Na₂O—MgO—SiO₂ has been studied by Russian investigators, who allege among other things a compound Na₂O·2MgO·6SiO₂. We have prepared a sample of this composition and have found that it does not behave as a compound.

CO-OPERATIVE INVESTIGATIONS

Age of minerals. An age-determination program (Davis, Nicolaysen, Aldrich, Tilton) was initiated last year in collaboration with the Department of Terrestrial Magnetism. The immediate purpose of the study is to develop techniques and modify equipment for determining the age of several common minerals, in the same rock, using different methods which will give completely independent ages. For example, work is under way on rubidiumstrontium, potassium-calcium, potassiumargon, and uranium-lead decay systems. Granite has been chosen as the most promising rock type for investigation because of its wide distribution throughout the areas in which Pre-Cambrian rocks are exposed. The mineralogical composition of granites is also well suited to the program;

the following minerals of granites all are possible candidates for age study: biotite, muscovite, allanite, apatite, ilmenite, magnetite, monazite, rutile, titanite, xenotime, and zircon.

Age-determination studies have many objectives. Perhaps the foremost goal is to gather new data bearing on the long-standing and classic problem of the earth's age. Information on this controversial subject will be of interest to astronomers, geologists, and physicists alike. The age of the earth is *in ambiguo*, and as science sharpens the tools for age determination, the earth's birthday is, year by year, being moved backward. At the beginning of the present century the earth was believed to be very young, 20 to 90 million years old. A few years later the age estimate was in-

creased to 2000 million years. At present the oldest minerals appear to be about 3500 million years old, and the age of the earth is believed to lie between 3500 and 5300 million years.

Stratigraphy of the great areas of crystalline rocks is another basic problem, where age determinations may serve to decipher the relationships, and where the classic methods of geology have failed to provide unambiguous answers. An example of this type of problem is the age of the great belt of crystalline rocks lying to the east of the Appalachian mountains. The stratigraphic position of this area has long been a controversial topic among geologists. One school maintains that these rocks are of Pre-Cambrian age, and a second group asserts that they represent metamorphosed Paleozoic rocks.

The origin of continents by gradual growth about a nucleus—in the case of North America, the Canadian shield region—is a fascinating hypothesis that may well stand or fall on the determination of the ages of Pre-Cambrian rocks throughout North America.

Knowledge of the relative ages of intrusive rock formations is of vital importance in many mining areas of the world. The locating of mineralized zones may depend upon knowledge of the genetic and structural relations between intrusive igneous rocks and the ore deposits. Thus, age studies may well have economic as well as scientific value.

In the past year efforts have been directed toward establishing reliable methods of age determination for minerals. Isotopic distribution in the elements of minerals is the primary source of data. The principal type of information utilized for age studies is gained from the naturally occurring radioactive elements having long half-lives. The problem is to separate, and measure quantitatively, the relative amounts of

parent and daughter isotopes. An example of the type of problem encountered is the measurement of the relative amounts of uranium, thorium, and lead in a granite where the concentration of uranium may be only 3 to 4 parts per million and that of lead 10 to 15 parts per million. Obviously the standard gravimetric methods of analysis are impractical.

The isotopic dilution method of analysis has proved uniquely suited to this problem. A known amount of a stable isotope is added to a known weight of a mineral, and the whole is decomposed by an appropriate procedure. When complete decomposition is attained, the added isotope dilutes or changes the isotope ratio of the element present in the mineral. All of the element in the resulting solution has the same isotopic composition, and separation and isotope analysis by mass spectrometer of any part of the solution will enable the absolute concentration in the sample to be determined with an error of only a few per cent. This method has been applied to the analysis for rubidium and strontium of lepidolite, biotite, and muscovite micas as well as of feldspars. Some preliminary work has been carried out on calcium; and zircon from granites has been analyzed for uranium, thorium, and lead.

Analysis of a mineral involves crushing, grinding, and sizing, with magnetic and density separations to free the chosen mineral of impurities. Granites are separated into the component minerals biotite, muscovite, feldspars, apatite, zircon, and so on. The final concentrate is "spiked" with a tracer isotope, then decomposed, and the desired elements are isolated for mass-spectroscopic analysis. This final separation of elements is effected by means of columns of ion-exchange resins with reagents that preferentially complex certain elements. This procedure, in effect, enables us to pluck out, from large quantities of ele-

ments of groups I and II, microgram amounts of potassium, rubidium, calcium, and strontium.

An example of the use of ion-exchange resins illustrates this separation. At a pH of 4.5, iron, aluminum, titanium, zirconium, manganese, and so forth, pass through a column of resin in the ammonia form when complexed with glycolic acid. The potassium and rubidium can then be collected and partially separated, leaving the calcium and strontium on the column. These can then be collected, and also partially separated. The separation of rubidium and strontium is complete, and the procedure is reproducible with no contamination of the samples.

Thus far, actual age determinations by rubidium-strontium ratio have been confined to lepidolite micas. The results are: Ingersoll Mine, South Dakota, 2100 million years; Winnipeg River, Manitoba, 3500; Bikita Quarry, Southern Rhodesia, 3300; Pala, California, 157; Jakkalswater, South Africa, 2400.

Spectrographic analyses of micas (Nicolaysen). The use of the optical spectrograph for quantitative determination of strontium and rubidium in micas has been investigated during the past year. Results indicate that selective volatilization of the fluorides prepared from micas introduces a serious difficulty. This factor, and the variations in chemical composition within the mica group, are apparently responsible for the lack of reproducibility. Examination of the "synthetic standard" fluorides and fluorides resulting from the decomposition of natural micas by X-ray diffraction showed greatly different powder patterns.

This indicates that variation in the crystalline structures formed in the two cases may be responsible for the differences in general arcing quality of the "synthetic standard" and "natural" specimens, introducing a systematic error in spectrographic age determinations.

In the course of this study it has been noted that large quantities of rubidium are present in all manganese-bearing pegmatite micas, and not merely in the lithium-rich lepidolites. This observation should enlarge the scope of precise age measurements on pegmatites.

Deep seismic prospecting. Further investigations by seismic methods have yielded results of interest and importance regarding the nature of the earth's crust. New field studies in Minnesota, in the state of Washington, and in other places have shown how the thickness of the crust varies from place to place on this continent. As might have been expected for other reasons, the depth at which the velocity of an elastic wave changes to the high value characteristic of the subcrustal (presumably olivinitic) material is greatest in places where the land masses are more elevated. The remarkable discontinuity that marks the bottom of the crust is at some places exceedingly sharp, but at other places it seems to be merely a gradual transition in wave velocity. Alternatively, the region, though described as a surface, may be rough and irregular. Details of this joint program of the Department of Terrestrial Magnetism and the Geophysical Laboratory will be found in the report from the former Department.

Listed below under "Summary of Published Work" are papers published during the report year in technical journals. Additional papers which have been prepared

and are awaiting publication are as follows: L. H. Adams, "A note on the stability of jadeite"; L. H. Adams, "List of systems investigated at Geophysical Labo-

ratory"; L. H. Adams and F. A. Rowe, "The preparation of specimens for the focusing type X-ray spectrometer"; Tom. F. W. Barth, "The differentiation of a composite aplite from the Pribilof Islands, Alaska"; A. F. Buddington, "Chemical petrology of some metamorphosed Adirondack gabbroic, syenitic, and quartz syenitic rocks"; F. Chayes, "A provisional reclassification of granite"; F. Chayes, "Relations between composition and indices of refraction in natural plagioclase"; G. L. Davis and O. F. Tuttle, "Two new crystalline phases of the anorthite composition, CaO. Al₂O₃·2SiO₂"; Gabrielle and J. D. H. Donnay, "The symmetry change in the high-temperature alkali-feldspar series"; P. Eskola, "On the granulites of Lapland"; H. W. Fairbairn and J. F. Schairer, "A test of the accuracy of chemical analysis of silicate rocks"; H. H. Hess, "Orthopyroxenes of the Bushveld type, ion substitutions, and changes in unit cell dimensions"; E. Ingerson, "Twinning frequency in feldspar phenocrysts from a quartz latite sill at Sierra Blanca, Texas"; M. L. Keith and O. F. Tuttle, "Significance of variation in the high-low inversion of quartz"; S. Kozu, "Japanese twins of quartz"; F. C. Kracek and K. J. Neuvonen, "Thermochemistry of plagioclase and alkali feldspars"; W. S. MacKenzie, "The effect of temperature on the symmetry of high temperature soda-rich feldspars"; G. W. Morey, "Flow of glass at room temperatures"; G. W. Morey, "Hydrothermal synthesis"; G. W. Morey, "The system sodium metaphosphate—calcium metaphosphate"; G. W. Morey and J. M. Hesselgesser, "The system H₂O-Na₂O-SiO₂ at 400° C"; K. J. Neuvonen, "Heat of formation of

merwinite and monticellite"; P. Niggli, "The chemistry of the Keweenawan lavas"; E. F. Osborn and D. B. Tait, "The system diopside—forsterite—anorthite"; E. Roedder, "A reconnaissance of liquidus relations in the system K₂O·2SiO₂—FeO— SiO2"; Th. G. Sahama, "Leucite, potash nepheline, and clinopyroxene from volcanic lavas from southwestern Uganda and adjoining Belgian Congo"; J. F. Schairer, "The system K₂O—MgO—Al₂O₃—SiO₂. I. Results of quenching experiments on four joins in the tetrahedron cordierite forsterite—leucite—silica, and on the join cordierite-mullite-potash feldspar"; J. F. Schairer and K. Yagi, "The system FeO-Al₂O₃—SiO₂"; J. V. Smith, "Crystal structure of paracelsian"; J. V. Smith, "Singlecrystal measurements on paracelsian"; J. V. Smith, "Re-examination of the crystal structure of melilite"; J. V. Smith, "Two aids for the orientation of crystals in a precession camera"; R. B. Sosman, "Temperature scales and silicate research"; C. E. Tilley, "Some trends of basaltic magma in limestone syntexis"; G. Tunell, "The angle between the a-axis and the trace of the rhombic section on the {010}-pinacoid in the plagioclases"; G. Tunell and L. Pauling, "The atomic arrangements and bonds of the gold-silver ditellurides"; O. F. Tuttle, "Optical studies on alkali feldspars"; K. Yagi, "Petrochemical studies on the alkalic rocks of the Morotu District, Sakhalin"; H. S. Yoder, "Change of melting point of diopside with pressure"; H. S. Yoder, "The 10 per cent CaAl₂Si₂O₈ plane in the system CaSiO₃—Ca₂Al₂SiO₇ —NaAlSiO₄—CaAl₂Si₂O₃"; H. S. Yoder, "The MgO—Al₂O₃—SiO₂—H₂O system and the related metamorphic facies."

SUMMARY OF PUBLISHED WORK

(1144) Jadeite of San Benito County, California: Preliminary report. H. S. Yoder and C. W. Chesterman. California Division of Mines, Spec. Rept. 10-C, 8 pp. (Sept. 1951).

The rare mineral jadeite, NaAl(SiO₃)₂, which is important to the general study of "high-pressure minerals," was recently discovered in California. Eight large exposures of jadeite were found in the canyon of Clear Creek, and there are, no doubt, many more. Six of these bodies, completely surrounded by highly sheared and brecciated serpentine, are lens-shaped. The seventh, a veinlike body, was found at the contact of the serpentine with an enclosed mass of schist. The existence of an eighth body is inferred from the large masses of float at the contact of another enclosed block of schist with the serpentine and from the presence of jadeite in the schist near the contact.

The lens-shaped bodies were composed of thinly banded, dark green jadeite in which were enclosed blobs of natrolite, pectolite, low-temperature albite, knots of black talcous material, and small irregular masses of thickly banded shaly rock. The contact zone, where observed, consisted mainly of grossularite, lawsonite, pumpellyite, and a green amphibole.

The veinlike body was traceable for about 50 feet and appears to be terminated at one end by large masses of jadeite similar to that of the lens-shaped bodies. The vein is mostly white jadeite, exceptionally rich in the jadeite molecule, with analcite and some low-temperature albite. The occurrence of jadeite in a vein associated with serpentine supports the view that jadeite does not require extreme pressures for its formation and that it is stable at moderate temperatures (less than 500° C).

(1145) Change of free energy with pressure of the reaction nepheline + albite = 2 jadeite. H. S. Yoder and C. E. Weir. Amer. Jour. Sci., vol. 249, pp. 683–694 (1951).

The change of free energy with pressure for the reaction nepheline + albite = 2

jadeite was computed from values of the density and compressibility determined by the writers and from the unpublished thermal expansion data of Rosenholtz and Smith, and was found to be very small.

On the basis of the thermodynamic values obtained by Kelley and by Kracek and Neuvonen it can be stated with certainty that jadeite does not *require* pressure for its formation at room temperature. The present calculations show that pressure *favors* the formation of jadeite, but at a slowly decreasing rate.

(1146) Computation of the optic axial angle from the three principal refractive indices. F. E. Wright. Amer. Mineralogist, vol. 36, pp. 543-556 (1951).

The standard equation for computing the value of the optic axial angle, $2V_{\gamma}$, of a biaxial crystal from its three principal refractive indices, α , β , γ , is: $\tan^2 V_{\gamma} = (1/\alpha^2 - 1/\beta^2)/$ $(1/\beta^2 - 1/\gamma^2)$. This equation is not in convenient form for use in computation. However, an approximate equation, $\cos 2V_{\sim} =$ $(\varepsilon - \delta)/(\varepsilon + \delta) - 6\varepsilon\delta/(\varepsilon + \delta)(\gamma + \alpha)$, in which $\varepsilon = \gamma - \beta$, $\delta = \beta - \alpha$, can be derived from it which is satisfactory and yields values accurate, in general, to 1 minute of arc for $2V_{\gamma}$, if the birefringence, $\gamma - \alpha$, does not exceed 0.050; and to 3 minutes of arc if $\gamma - \alpha$ is between 0.050 and 0.100. The simplified formula shows clearly that the optic axial angle is primarily dependent on the difference between the partial birefringences, $\gamma - \beta$ and $\beta - \alpha$, divided by the maximum birefringence, $\gamma - \alpha$, rather than upon the actual values of α , β , γ . The size of the index ellipsoid itself depends on the values of the principal refractive indices; its shape, on the other hand, depends on relations between the principal birefringences.

(1147) Complete substitution of aluminum for silicon: The system 3MnO·Al₂O₃·3SiO₂
—3Y₂O₃·5Al₂O₃. H. S. Yoder and M. L. Keith. Amer. Mineralogist, vol. 36, pp. 519–533 (1951).

Thermal, optical, and X-ray data indicate that there is a complete solid solution series between spessartite, $Mn_3Al_2(SiO_4)_3$, and yttrogarnet, $Y_3Al_2(AlO_4)_3$. The substitution $Y^{+3}Al^{+3} \rightarrow Mn^{+2}Si^{+4}$ may be represented by the formula $(Mn_{3-x}Y_x)Al_2(Si_{3-x}Al_x)O_{12}$.

Yttrogarnet has a cube edge of 12.01 \pm 0.02 Å and the space group O_h^{10} . Using the co-ordinates of grossularite as determined by Menzer, good agreement was obtained between calculated and observed intensities. The atomic positions are as follows:

Al in 16(a) 0 0 0
Y in 24(c)
$$\frac{1}{4}$$
 $\frac{1}{8}$ 0
Al in 24(d) $\frac{1}{4}$ $\frac{3}{8}$ 0
O in 96(h) x y z

where x, y, and z are 0.04, 0.055, and 0.64, respectively. The yttrium is in eightfold coordination; the aluminum is in both four- and sixfold co-ordination. Yttrogarnet inverts to a high form, yttroalumite, at 1970° \pm 50° C.

This is the first case on record of the complete substitution of aluminum for silicon in an orthosilicate. The complete solid-solution series contradicts the prevalent idea that the substitution of aluminum for silicon always decreases as the Si/O ratio decreases.

(1148) A test of the precision of thin-section analysis by point counter. F. Chayes and H. W. Fairbairn. Amer. Mineralogist, vol. 36, pp. 704–712 (1951).

Five thin sections from Westerly, Rhode Island were analyzed by each of five relatively inexperienced operators using the pointcounter method of determining the mode. Each operator ran his slides in a different sequence. In a preliminary run the observed analytical error for the constituents exceeded the expected error; in the second run by the same operators, now more experienced, the observed analytical error was approximately the same as the expected error. Evaluation of variances showed order of analysis to be insignificant throughout, and operator and slide differences to be slight. The means for all constituents were remarkably close to the means previously obtained by Chayes in an independent run on the same sections. The test definitely establishes the level of precision for the point-counter method of analysis on a broader base than was possible from Chayes' unchecked analyses. The convenience and rapidity of the method are already well known; its precision has now been thoroughly tested.

(1149) Double fluorides of zinc. E. Ingerson and G. W. Morey. Amer. Mineralogist, vol. 36, pp. 778–780 (1951).

The preparation of the compounds ZnF·4H₂O, NH₄F·ZnF₂, MgF₂·ZnF₂, CaF₂·ZnF₂, SrF₂·ZnF₂, BaF₂·ZnF₂, MnF₂·ZnF₂, and ZnF₂·ZnCl₂ is described, and melting points and optical properties are given for them as well as for ZnF₂.

(1150) Phase transformations in one-component silicate systems. F. C. Kracek. Reprinted from Phase transformations in solids, by Smoluchowski, Mayer, and Weyl, pp. 257–277. New York, John Wiley & Sons (1951).

A discussion is given of the characteristics of melting, crystallization, and polymorphic transformations in silica and in silicate compounds considered as one-component systems.

The well known property of silica molecules of polymerizing in extended networks of linked SiO₄ tetrahedral groups greatly influences the speed of phase transformations in these compounds and leads to the existence of metastable phases at temperatures and pressures far removed from the equilibrium transformation points. The solidification of these substances as glasses, the difficulties of devitrification, and the existence of metastable modifications in nature as minerals are examples of sluggish transformation.

In a consideration of the reversible polymorphic transformations in silica and silicates, two principal cases must be distinguished; namely, the sluggish and the rapid transitions, illustrated by the quartz-tridymite and quartz-cristobalite or wollastonite-pseudo-wollastonite inversions on the one hand, and the high-

low quartz or leucite transitions on the other. Structural evidence indicates that a rearrangement of bonds and a major change of symmetry take place in what we call a sluggish transition, whereas in the rapid type the change of symmetry is only minor and involves no breaking of bonds. The two types have been called the reconstructive and displacive transitions, respectively, by Buerger.

Examples of the sluggish and rapid types of transition are given; no pretension is made to completeness of treatment or exhaustiveness of literature citations, as the examples were chosen largely for the purpose of indicating the types of behavior encountered among the silicates. Quantitative data on the energy and volume relations are available for only a few of the transitions mentioned. Data are also needed on the stability relations of the monotropic forms of silicates encountered in nature. Good examples of these are the three modifications of Al₂SiO₅, sillimanite, and alusite, and kyanite.

(1151) Phase transformations in polycomponent silicate systems. J. F. Schairer. Reprinted from Phase transformations in solids, by Smoluchowski, Mayer, and Weyl, pp. 278–295. New York, John Wiley & Sons (1951).

Phase transformations in polycomponent silicate systems are discussed, including melting, crystallization, solid solution, and changes in crystal form and composition. The phenomena of congruent and incongruent melting and the relation of solid solution to these phenomena and to inversions in solid phases are presented. Phase-equilibrium diagrams depict the stability of the several single or coexisting solid phases. The effect of solid solution on inversion is discussed in some detail. The evidence for homogeneous equilibrium in the liquid above the congruent melting point of albite is presented here for the first time, and its effect on crystallization is emphasized. The need for closer co-operation between physicists, physical chemists, metallurgists, and ceramists, and the need for a uniform nomenclature are stressed.

- (1152) Annual Report of the Director for 1950–1951.
- (1153) The ternary system Na₂O—B₂O₃—SiO₂. G. W. Morey. Jour. Soc. Glass Technol., vol. 35, pp. 270–283 (1951).

In this study of the phase equilibrium relations in the system Na₂O—B₂O₃—SiO₂, the fields of the compounds have been outlined and isotherms determined. Portions of the system are exceedingly difficult to crystallize, and no mixture in the binary system B₂O₃—SiO₂ has been crystallized. A new compound, Na₂O·B₂O₃·2SiO₂, has been obtained, and the conditions for its preparation and its melting point have been determined, but its field and its relation to the field of the other compounds have not been learned.

(1154) The solubility of quartz and some other substances in superheated steam at high pressures. G. W. Morey and J. M. Hesselgesser. Trans. Amer. Soc. Mech. Engrs., pp. 865–875 (Oct. 1951).

An apparatus and method for determining the solubility of silica in superheated steam at high pressures are described. At 752° F (400° C) the solubility of quartz in steam increases from 1.0 ppm at 500 psi to 1548 ppm at 15,000 psi; at 932 F (500° C), the solubility increases from 4.3 ppm at 500 psi to 2596 ppm at 15,000 psi. The curves representing the solubility results at 752° F and 932° F cross at two pressures. Between these two pressures the solubility is greater at 752° F than at 932° F; below and above these pressures the solubility is greater at 932° F than at 752° F. This crossing is ascribed to the pressure-density relations of water at the two pressures, which are affected by the proximity at 752° F of the critical point of water. When the solubility is plotted against the density of steam at each measured point, the curves do not cross, and the solubility at 932° F is greater than at 752° F. The solubilities of some other substances in superheated steam at high pressures are also given.

(1155) Elastic properties of materials of the earth's crust. L. H. Adams. Reprinted from Physics of the earth—VII, Internal constitution of the earth, edited by Beno Gutenberg (2d ed.), ch. 4, pp. 50–80. New York, McGraw-Hill Book Co. (1951).

This chapter is from the new edition of the book first published in 1939, and is a revised and restated summary of this subject (Papers from the Geophysical Laboratory, No. 1007). A useful table is the one giving newly calculated velocities of longitudinal and transverse waves through various types of rock at different depths within the earth's crust.

(1156) The chemical and petrological nature of the earth's crust. H. S. Washington and L. H. Adams. *Reprinted from* Physics of the earth—VII, Internal constitution of the earth, edited by Beno Gutenberg (2d ed.), ch. 5, pp. 81–106. New York, McGraw-Hill Book Co. (1951).

This is a revision and condensation of the chapter entitled "The crust of the earth and its relation to the interior" (Papers from the Geophysical Laboratory No. 1008) in the earlier edition of the book. An important addition is a diagram showing the composition, in terms of the minerals, of the important rock types.

(1157) On supersonic flow past a slightly yawing cone. II. A. H. Stone. Jour. Math. and Phys., vol. 30, no. 4, pp. 200-213 (1952).

This investigation, like that described in the first paper of the series (Papers from the Geophysical Laboratory No. 1101), was carried out for the Office of Scientific Research and Development as a war problem. The present paper continues the analysis of the flow produced by a circular cone of not too blunt an angle moving through air at high speed in a direction making a small angle with its axis. The object of the investigation described here was to carry the relations to a

higher degree of approximation. The method is essentially the same as was followed in the previous communication. Because the fundamental equations are nonlinear, a complication arises in that the flow exhibits some unexpected singularities. Various observations of conical projections in flight show a satisfactory agreement with the theoretical equations.

(1158) Thermochemistry of mineral substances. I. A thermodynamic study of the stability of jadeite. F. C. Kracek, K. J. Neuvonen, and G. Burley. Jour. Wash. Acad. Sci., vol. 41, pp. 373–383 (1951).

The tendency to form jadeite has been determined by evaluating the standard free energy changes $\Delta F^0_{298.16}$, at 1 atm for the reactions

- A. Albite = jadeite + quartz
- B. Albite + nepheline = 2 jadeite
- C. Nepheline + 2 quartz = albite
- D. Nepheline + quartz = jadeite

The values of $\Delta F^0_{298.16}$ were obtained by the relation $\Delta F^0_{298.16} = \Delta H^0_{298.16} - T\Delta S^0_{298.16}$ from entropies furnished by K. K. Kelley in a personal communication and from the standard enthalpy changes $\Delta H^0_{298.16}$ for the reaction, as derived from measurements of the heats of solution of the reaction constituents in 20 per cent hydrofluoric acid in a calorimeter operated at 74.7° C, which is described.

The influence of the application of high pressures is evaluated from available exact measurements of the densities and compressibilities of the reaction constituents. The results of the calculations indicate that pressure slightly favors the advance of reactions A, B, and D, as written, from left to right, whereas for reaction C its effect is in the reverse direction and negligibly small.

The results indicate that a tendency exists to form jadeite at 25° by reactions B and D at 1 atm pressure, and that this tendency is increased by the application of high pressure; and, further, that by reaction A jadeite be-

comes stable only at pressures beyond 1600 bars.

(1159) The solubility of some minerals in superheated steam at high pressures. G. W. Morey and J. M. Hesselgesser. Econ. Geol., vol. 46, pp. 821-835 (1951).

The solubility of salts and minerals in superheated steam at high pressure is of importance to geologists in their attempts to interpret and explain the genesis of pneumatolytic and vein deposits, and the experimental demonstration and measurement of such solubilities is of interest to them. The present paper describes an apparatus and a method which have been used for this purpose at temperatures up to 600° C and at pressures up to 2000 bars.

(1160) Notes on the staining of potash feldspar with sodium cobaltinitrite in thin section. F. Chayes. Amer. Mineralogist, vol. 37, pp. 337-340 (1952).

Several simplifying modifications of the staining procedure are described. The behavior of soda-rich or minutely perthitic potash feldspar is anomalous, but the stain is highly selective for microcline in the presence of plagioclase and will not obscure perthitic intergrowths coarse enough to be readily resolved in unstained thin sections. The stain greatly facilitates both modal analysis and ordinary petrographic examination.

(1161) Charts for the interpretation of rotation and Weissenberg photographs. Gabrielle Donnay and J. D. H. Donnay. Rev. Sci. Instr., vol. 23, pp. 130-132 (1952).

Two charts are presented. The first one gives, from a rotation photograph, the direct lattice translation of the rotation axis. From a zero-layer undistorted Weissenberg photograph, the second chart gives the reciprocal lattice translations perpendicular to the rotation axis. The nomographs are constructed for the standard camera radius (R=180/2 mm), but they can be used with any wave length.

(1162) The stability field of sapphirine in the system MgO—Al₂O₃—SiO₂. M. L. Keith and J. F. Schairer. Jour. Geol., vol. 60, pp. 181–186 (1952).

A detailed study, by the method of quenching, of ten closely spaced compositions in a small portion of the ternary system MgO—Al₂O₃—SiO₂ has delineated the tiny field of sapphirine and located the temperatures and compositions of three ternary reaction points. Sapphirine crystals are stable in equilibrium with a liquid in the ternary system only over the narrow range of temperature between 1482° and 1453° C.

(1163) Origin of the contrasting mineralogy of extrusive and plutonic salic rocks. O. F. Tuttle. Jour. Geol., vol. 60, pp. 107–124 (1952).

Mineralogical differences between the quartz and feldspars of extrusive and plutonic acidic rocks are believed to represent changes brought about in the minerals of plutonic rocks by inversion, unmixing, and recrystallization from an initial character similar to that of the quartz and feldspars of the corresponding extrusive rocks.

Equilibrium studies in the system orthoclase—albite—water provide quantitative information on the stability relations of the alkali feldspars. A single alkali feldspar is stable at high temperatures, but this phase becomes unstable at low temperatures and breaks up into a potash-rich and a soda-rich phase (perthite). Such unmixing is common in the perthite-quartz granites. Granites of orthoclase—plagioclase—quartz (no perthite) may have the same chemical composition as the perthite-quartz granites, and it is suggested that the unmixing process may continue beyond the perthite stage to give nearly potash-free plagioclase and soda-free orthoclase. Such a process of unmixing and recrystallization may explain conflicting evidence on the order of crystallization in granitic rocks. It is believed that these processes, the result of changing stability relations at low temperature, have produced significant

changes in the mineralogy and textural relations of the plutonic acidic rocks.

(1164) On the association of perthitic microcline with highly undulant or granular quartz in some calcalkaline granites. F. Chayes. Amer. Jour. Sci., vol. 250, pp. 281–296 (1952).

Microperthite is on the whole uncommon in the micaceous calcalkaline granites of New England, Texas, and the southeastern states. It is usually rare or lacking if quartz is ungranulated and shows only moderate undulance; it may or may not be common if quartz extinction is highly undulant, and it is never rare if quartz is extensively granulated. The shearing stresses which render quartz extinction undulant and finally granulate the quartz evidently induce or accelerate the unmixing of albite-poor microcline, which might otherwise remain indefinitely metastable. Alternatively, if the albite is already submicroscopically exsolved, as often seems to be the case, stress promotes its recrystallization into units of optically resolvable dimensions.

(1165) Radioactivity of ocean sediments. VIII.
Radium and uranium content of ocean
and river waters. Elizabeth Rona and William D. Urry. Amer. Jour. Sci., vol. 250,
pp. 241–262 (1952).

New determinations of the radium and uranium content of ocean and river waters are reported here, and it appears possible to draw some tentative general conclusions concerning the distribution of the radioelements of the uranium 238 series in the hydrosphere from these measurements and the few reliable determinations reported in the literature. The conclusions are based on a few data and are presented to indicate possible fruitful lines of future endeavor in this field; they should not be construed as forming a completed general theory.

Variations of the radium content of ocean

waters, both laterally and in depth, appear to be attributable to the presence of different water masses. Open ocean waters contain only about 16 per cent of the amount of radium that would be in radioactive equilibrium with the uranium content, a finding in agreement with the work of previous investigators on inshore waters. This departure from equilibrium is quantitatively the complement of the departure from equilibrium in ocean sediments now being deposited. Ocean waters lack also the ionium content required for equilibrium with the uranium content. A few measurements of the radium and uranium content of river waters show that the departures from radioactive equilibrium are in the opposite direction from similar departures in ocean waters, and in this respect the relations between the radioelements in river waters are qualitatively the same as those in ocean sediments.

(1166) The finer-grained calcalkaline granites of New England. F. Chayes. Jour. Geol., vol. 60, pp. 207–254 (1952).

Modal analyses for a collection of 145 thin sections of New England calcalkaline granites are presented and discussed. The data are used to estimate a range within which average or bulk modal compositions may be expected to fall. The internal variability of such granites is also examined in some detail. Remarkable internal homogeneity, rather small differences between granite masses, and consistent relation of data points for each mass to the thermal valley in the system quartz-orthoclase-albite all suggest the existence of granitic (liquid) magmas as parent materials. The origin of these magmas is not known; the most likely processes appear to be selective re-fusion and crystal fractionation. Critical evidence permitting a choice between these alternatives is not available, and it is curious that the difficulties confronting them are nearly identical.

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DEPARTMENT OF TERRESTRIAL MAGNETISM

Washington, D. C.

MERLE A. TUVE, Director

INTRODUCTION

The terms "natural philosophy" and "natural history" served to characterize the broad subdivisions of science a hundred years ago, and gave some indication, as well, of the general motivations of those who dedicated their efforts to the increase and diffusion of knowledge in these fields. The undeniably real pressures for specialization, as the range and intensity of the intellectual effort required for mastery of any subject kept mounting in accelerated tempo, have led in recent decades to some grotesque examples of the scientific specialist as one who knows more and more about less and less.

Even the progress of research in science has suffered distorted emphasis by reason of the narrowness of competence and interest exhibited by the specialized investigator. Yet today, more than ever before, both the opportunity and the necessity for a wide range of sympathetic interest and comprehension on the part of the scientific man has become clear, as the developments of science, often reduced to practice in technology or medicine, impinge on government policy and even on the moral decisions of each free citizen, in the support or opposition he gives to the kinds of manipulation which modern technology and health procedures impose on human beings and their relations to one another.

It is interesting to observe a growing recognition of this situation by some of the outstanding younger research men during the past decade or so, and especially since the war. Two features are characteristic, namely, recognition that fruitfulness in

scientific work requires alert comprehension of progress occurring in several or many fields, not just in one narrow specialty, and recognition that the motivation of a research man or a research program is a major factor in characterizing the fruits of the program.

The interested but dispassionate seeker after new knowledge or new aspects of the magnificent orderliness of nature, the worker in pure science, shares the traditional motivation of the natural philosopher. The opportunity for that kind of dedicated effort is not often matched to the readiness of the individual, but one of the very special ways in which the Institution differs from other research agencies in this postwar world, with incredible sums being spent yearly for various kinds and qualities of research (and engineering), lies in the freedom of the staff and the departments to work on problems of their own selection. A primary criterion is the individual investigator's own estimate of the philosophical significance of a proposed research task, and the selection or emphasis among research problems is not affected by voters or stockholders or even by the legitimate needs of students in training.

That this special characteristic and opportunity of the Institution is fully grasped by the members of this Department, as a group of physicists each with compelling interests of his own, but keenly aware of the important steps being taken in many other fields, is amply illustrated by the wide range of special interests covered in the list of active investigations. It is also encouraging to discover, by actual test, that it still is feasible, in a modestly effective but satisfying way, for a specialized investigator to be aware of the important and current developments in many fields. Most of the investigations in progress involve participation by several staff members. These range from the time scales of Pre-

Cambrian geology and the evolution of continental structures through radio propagation, cosmic rays, and nuclear physics to the dynamics of the incorporation of amino acids into proteins, but each investigator remains characteristically a physicist, with the ancient motivations and satisfactions, within his own limits, of the "natural philosopher."

EXPERIMENTAL GEOPHYSICS

THE EARTH'S CRUST

The broad objective of the group concerned with this area of the Department's activities is to enlarge our understanding of the early history of the earth. We seek to enrich the significance of our questioning about the earth as an object in the solar system and as a whirling, heated globe on which mountains, seas, and continents rose and subsided during the two billion years or more which passed by before fossils were first laid down in the Cambrian period. The early history of life on the earth is an integral part of this same great panorama, and it may prove possible to find evidence of early living forms in tracemetal residuals or in isotope ratio differences in the ancient sedimentary rocks laid down before the bony structures of animals began to leave their imprints as fossils. The direct effect of life on the weathering of rocks and other geological phenomena is not small, because the oxygen and much of the nitrogen of the atmosphere is freed and maintained by life processes which were operating for a very long time before the Cambrian period. The character of the earliest atmosphere is a matter of complicated speculation, but a chemical record may lie hidden in the Pre-Cambrian rocks.

It is interesting to the members of this group, as physicists, to explore the many ways in which the ideas and techniques of modern physics and chemistry can bring to light new facts about the early history

of the earth. One expression of this interest lies in the phrase "evolution of continental structures," which illustrates the character of the questions under study. It is not clear in this decade whether the earth was formed from a ball of flaming gas torn from the sun, or by the gravitational aggregation of countless millions of small cold lumps of matter, heating after they collided and aggregated, in part by gravitational energy and in part by the radioactive energy evolved in a subsequent billion years or more. The end result in either case might be the earth as we know it today, with a heavy core, a mantle, and a crust.

The nature of the differences, seemingly original and permanent, which differentiate continental areas from deep ocean areas is a part of this general problem which calls for detailed observation and study. Questions relating to the forces which give rise to folding and mountain building, and the dating of the ancient epochs which have left residual geological structures as mute evidence of vast tectonic activities, are among the old problems to the solution of which significant contributions can perhaps be made by newer methods.

Modern physics and chemistry, by isotope and radioactivity determinations, often with microgram amounts of material, offer a new opportunity for the dating of igneous intrusives. There is evidence indicating that certain selected crystalline mineral

constituents are free from the effects of solution or redeposition that might have occurred after the initial igneous activity; these can be separated, for example, from granite. It may also be possible by modern technical procedures, after some years of comprehensive effort, to give answers to the question whether or not the growth of continents has been by accretion, that is, by the formation and erosion of successive mountain structures at the continental margins.

If we are to understand how and why the structure of continents differs in some basic and permanent fashion from the structure of deep ocean basins, or to know in what sense or in what ways this is not true, it is necessary to make observations on the underlying structures beneath the continents and the deep oceans. The developments of seismology and geophysical prospecting make it feasible to examine the crustal structures down to depths of 50 km or more, by observations on the waves from large explosions. Correlating this type of information with the data from gravity measurements should clarify many of the older uncertainties regarding the uniformity of the crust of the earth and the evolution of its slowly changing external features.

A more subtle question, and probably a more deep-seated one, both as to the structure of the earth and as to the complex physics of internal motions in the core, concerns the pattern of the magnetic field of the earth during past geologic time. Study of the sediments over a time range of a few thousand years in glacial clays, and in the Paleozoic rocks back to 300 million years, discloses a striking pattern of uniformity in the traces of residual magnetism and magnetizability. Whether this indicates that the magnetic field of the earth has been roughly constant during this long period remains uncertain. The crucial

studies relate, of course, to the degree of magnetic stability of the rocks, and this question is being examined in folded and flat-lying sediments, and in many ancient conglomerates.

Although the individual studies may seem technical and highly specialized, these brief paragraphs may have indicated the perspective in which the technical work is placed by the men who are so vigorously interested in the light which modern laboratory procedures can shed on those distant epochs before the Cambrian period.

SEISMIC WAVES FROM EXPLOSIONS

H. E. Tatel, M. A. Tuve; collaborators, L. H. Adams, J. W. Greig, P. A. Scherer

During the past five years the seismic waves at distances up to roughly a hundred miles from more than two hundred large explosions have been observed in field work by the Geophysical Laboratory and this Department in the eastern United States, New Mexico, and California. The measurements have sought quantitative data on the basement rocks underlying the sedimentary layers to a depth of 50 km or more. The observations have shown that a sharp boundary exists, at least in many places, between the crustal rocks and the higher-velocity rocks of the mantle. This boundary is at a deeper level under the Appalachian mountains than under the coastal plain, and it is slanting or rough and broken in some localities; it was not observed in a long series of special observations in southern California. The textbook picture of a downward succession of several horizontal granitic and basaltic layers was not confirmed, even in special explosion experiments designed to make such structures prominent features of the records. Evidence was found, instead, for real inhomogeneities in the crustal rocks down to 30 km or more, as though they were irregular in vertical and horizontal dimensions, as the basement rocks appear when exposed at the surface.

This general pattern of exploration of the underlying structure of this continent by seismic surveys has been continued. At the close of the last report year we had completed observations in Minnesota. The provisional results have not been changed. We were able to observe large reflections from the lower crustal discontinuity, which established its existence under Minnesota. As in the eastern United States, this discontinuity is not observed at every point where it might be expected, but it is unmistakable when seen. We have come to believe that this irregularity in the reflection pattern, in the zones beyond 100 km from the shot point, is a manifestation of the unevenness or the topographic features of the discontinuity, which under Minnesota is 40-44 km deep.

A vigorous search for this discontinuity between crust and mantle was also carried out in the state of Washington through the co-operation of the Office of Naval Research and the Coast Guard, which arranged to have the USCG Cutter Mallow set off a series of charges in Puget Sound during July 1951. Our expedition had five observing groups in this area for more than two weeks. Three stations carried out observations using two-pen recording seismographs, and two stations used 24channel multiple recorders, with seismometers distributed at intervals along 3600foot cables. Observations were made throughout the state of Washington to the east and south, and west to the tip of Cape Flattery. The results were quite different from those which had been found just previously in Minnesota. Our major effort was to locate critical reflections from the lower crustal discontinuity, but we found no consistent pattern of pulses which would indicate a buried reflecting layer. There was evidence of a fairly deep sedi-

ment layer, and there was evidence that the velocity increased with depth. The velocity of the compressional waves in the upper crust is close to 6 km/sec, as elsewhere. One surprising observation was the finding of evidence for deep-lying highvelocity rock rising rapidly toward the surface westward from Seattle as one moves toward the Pacific. Usually the first "break" in the travel-time curves, indicating wave travel in rocks of higher velocity than near the surface, is at 140 km, and this is the case for the observation line to the east of Seattle. But to the west, the break is in evidence at 110 km, and if we had been able to obtain a greater number of observations nearer the shots, the records indicate that the break would have been observed as close as 80-90 km from the shot. This effect is much too large to be attributed to thinning of the sediments, and we take it as evidence that the high-velocity rock (8.1 km/sec) rapidly rises toward sea level as one approaches the west edge of the Olympic Peninsula.

Though the measurements in the vicinity of Puget Sound are too few to make extensive speculation possible, they are a good indication of structural complexities. In general, the results of these measurements in the state of Washington are similar to those made in southern California three years ago. There we found no system of reflections from the lower crustal boundary. The conclusion is that in both these areas the lower crust is different from that in the eastern part of the continent. The difference may arise from the lack of a discontinuity in these areas, or from the roughness of the discontinuity if it exists. There are similarities in the several regions of the United States we have explored in that the upper crustal rock velocity is near 6 km/sec for compressional waves and that it increases with depth. In the east it increases to slightly over 8 km/sec at depths

of from 32 km near the coast to about 48 km under the Appalachians. Near the west coast the measurements, because of scatter of the observations, due to real differences in travel times to specific observing points, are not good enough to give the velocity below the crustal material with any precision, but they are not discordant with a value of 8 km/sec for depths below roughly 45 km.

The multiple-trace seismograms have shown patterns which in some cases have unexpected features. During the first few seconds after the first arrival (at a distance of 100 km or so) all seismometers in a line 3600 feet long, in the direction of the shot, show nearly the same ground motion, with appropriate delays from one seismometer to the next. This means that the radiation is apparently coming in from the direction of the source. Later the motion becomes quite different for the various seismometers, showing side radiation and interference patterns randomly passing over the line of seismometers. Superposed on this pattern are what look like a series of recurrent waves of somewhat longer period. The origin of these ground motions, which seem to be a property of the region observed, is not understood.

We have constructed a portable displacement seismometer with period adjustable to up to 10 seconds. It has operated since December, and we have observed Love waves from California earthquakes. These were first observed at Columbia University; they are of interest to us because their dispersive properties and period depend upon crustal characteristics.

Tests were completed just as the report year closed (June 1952) in which an effort was made to observe reflections from the Mohorovičić discontinuity under the Great Plains, using the small explosions (5 to 20 pounds) fired daily by a seismic crew mapping structures in the search for oil. About

35 observations were made in three compass quadrants at distances of 110 to 190 km in Colorado and Nebraska from about 20 shots fired in southwestern Nebraska by a field party of the Magnolia Oil Company under the supervision of Mr. John Paige. We are grateful to Mr. C. G. Dahm, Dr. Milton Dobrin, and Mr. Paul Nash, of the Magnolia Company, for facilitating these tests. Good records had been obtained with the same instrument sensitivities on the Atlantic coastal plain at 130 km with shots of similar size, but the noise level over the deep sediments of the Great Plains was much higher, and the absorption evidently was also greater; no shots were observed on the seismometers despite numerous shifts to selected locations. Large explosions regrettably seem to be necessary for observing the deep structures under the Great Plains.

It is appropriate to mention in this report that a considerable fraction of the total staff time and effort of each group is spent in study and discussion of many other avenues of approach to the problems actually under investigation, primarily in order to formulate fresh questions and to recognize fresh areas of interest. For example, during this year new impetus has been given to our continuing interest in the development of a convenient and rapid gravity meter for work at sea (in submarines). This has been reinforced by the recognition that a detailed gravity survey over a restricted area of the deep sea may be a good way to determine whether or not the ultrabasic rocks of the earth's mantle, under the Mohorovičić discontinuity, are uniform (8.1 km/sec velocity), or are roughly similar to the inhomogeneous mixture we have found in the crust above the discontinuity. A few years ago the crust was pictured as a series of uniform layers, and today the deeper rocks are treated as uniform. The deep sea bottom is in many

areas now thought of as composed of this ultrabasic rock covered by a kilometer or two of mud. If a detailed gravity survey of an area the size of Minnesota over the deep sea should show localized gravity anomalies like those in the gravity map of Minnesota, for example, we could no longer picture the ultrabasic rock under the discontinuity as a uniform medium.

Similar discussions concerning heat flow, convection, and electric currents in the deep mantle and core have led to some simple formulations, especially in terms of thermoelectric currents and the Hall effect, which do not appear to have been suitably explored in relation to the continuing lack of a suitable theory of the earth's magnetic field. These discussions have led to the formulation of possible experimental tests of particular theoretical possibilities.

ISOTOPE DATING OF ANCIENT SEDIMENTS AND IGNEOUS INTRUSIVES

L. T. Aldrich, G. R. Tilton; collaborators, G. L. Davis, L. O. Nicolaysen

Although geological age measurements by uranium-lead ratios have been accumulating slowly since early in the century, when this procedure drastically revised all ideas of geological time scales, the total number of such measurements is not large. There also are reasons to distrust the claims of precision, because no way has been found for specifying the isotope ratio of the lead or uranium present when the earth was formed, and there is no way of knowing the degree to which subsequent mixing may or may not have occurred in the materials of a given ore body. There have been many efforts to weigh and to judge the available information, but it is clearly important to obtain independent measures of the ages of specific rock masses, using several different isotope methods. The long-period radioactivity of potassium and rubidium offer opportunities for checking the uranium-lead, thorium-lead, and lead-lead ages, especially if measurements on all these elements can be made at the microgram levels at which they are found, for example, in igneous formations.

In general, the study of isotope ratios in naturally occurring substances can give rise to two types of information. One is concerned with the parent-daughter relationship of radioactive decay. With suitable evidence as to the initial ratio when the rock was laid down (for example, if one constituent was practically excluded by the initial chemical crystallization), and with evidence that subsequent dilution or mixing did not occur, reliable determinations of the age of the mineral structure can be obtained. The second type of information relates to variations in isotope ratios when no radioactive decay is known to have taken place. In this case, isotope fractionation may have occurred, as in the passage of carbon through living plants, or by participation in some other metabolic process of a living organism; or varying isotope ratios may lead to the discovery of unsuspected radioactive instabilities. In recent years the possibility of isotope separation in very small degree by chemical processes has also been demonstrated.

In the program of the Department, broadly designed as a foundation for measurements on the ages of igneous intrusives in many Pre-Cambrian areas of tectonic activity, great progress has been made in demonstrating the reliability of special procedures for determining isotope ages of microconstituents of igneous rocks. The program is a co-operative one between staff members of the Geophysical Laboratory and staff members of this Department, and involves collaboration also with the Geology Department of the Massachusetts Institute of Technology (L. H. Ahrens, P. M.

Hurley, L. F. Herzog). The newer procedures, utilizing adsorption on ion-exchange resin columns and quantitative measurements of the absolute amounts of each chemical element by use of the isotope-dilution technique and the mass spectrometer, are reliable, readily tested at the microgram level, and convenient to carry out in a physics laboratory. The techniques are mastered by each investigator and cover every aspect of the technical procedure from crushing the original granite to the final mass-spectrometer operation. Preliminary studies were made utilizing spectrochemical determinations made elsewhere as part of the data, with isotope determinations made here, but later procedures involve making all the determinations by isotope dilutions and resin column procedures, without use of the optical spectrograph, and thus permit check measurements to exclude systematic errors. It is well known that the presence or absence of easily ionized elements can give rise to very troublesome corrections in spectrochemical determinations using the optical spectrograph, and the igneous rocks for which isotope dates are desired contain highly variable amounts of elements which can affect the arcing conditions for a spectrographic exposure.

On the basis of the experience of the past year in making and cross checking determinations of the amounts of the significant elements concerned in six or seven radioactive age ratios, it appears feasible to initiate a program of measurements concerned with the problem of continent formation. The oldest known part of the North American continent is in Manitoba, and much evidence seems to indicate that the continent has grown by successive increments from this vicinity. Field trips have therefore been organized for collecting suitable igneous rock samples from the southeastern, southwestern, and northwest-

ern United States, to look for minerals of great age in zones far from the Canadian shield, which may have been the ancient center of growth for the whole continent.

Age Measurements Using Potassium and Rubidium Elements

Experience in making actual age determinations on a series of selected minerals during the year has confirmed the initial choice of methods as being satisfactory. Known small amounts of stable isotopes, obtained from the Atomic Energy Commission, are used as tracers, which are added to the initial mineral sample and carried through all subsequent chemical procedures to the final measurements of quantitative ratios on the mass spectrometer. The tracer isotope intensity is given in micrograms by the known quantity initially added, and this calibrates the quantities of all other isotopes also in actual micrograms. The chemical determination of the parent element is made in the same fashion, and the two analyses on the mass spectrometer give all the data required for each age determination.

To avoid confusion from isotopes of different elements occupying the same position in the mass spectrometer, it is essential to carry out a reasonable degree of chemical purification. Techniques have been perfected during the year, with the help of the biophysics group, for extracting the trace amounts of calcium and strontium found as daughter elements in the potassium and rubidium minerals which we are using for age determinations. These new methods involve the use of ion-exchange resin columns, and certain special reagents which form complexes with specific elements when they are found in solution. By these procedures it is possible to separate microgram quantities of calcium and strontium from gram quantities of iron, aluminum, magnesium, and silicon. The

calcium and strontium thus obtained not only are free of these other elements, but also are well separated from each other. These procedures have been checked and cross checked on test samples, utilizing the isotope dilution technique, and the columns are found to be reproducible in their chemical behavior and to add no contamination to the samples prepared for measurement. The ease and precision with which an unknown mineral sample can be "spiked" with a measured quantity of a separated isotope, permitting quantitative determinations of the desired unknown. without correction for amounts which may be lost on the walls of beakers or in partial precipitation, is impressive and highly reassuring.

To date an igneous intrusive with some degree of reliability, it is important to measure its age by at least two independent radioactive decay products. Facilities and procedures have been worked out for using the Rb87-Sr87 and the K40-Ca40 natural decay schemes. During the year we have also participated in refining and checking the uranium-lead and thoriumlead age measurements on igneous mineral constituents, and facilities for complementary analyses by all these methods are now available. It is expected that the completion of the equipment for the K40-A⁴⁰ determinations will be carried out during the coming winter. The cross checking of mineral ages by use of several methods on the same rock should give reassuring confirmation or grave warnings about specific age determinations.

The actual measurements carried out this year were concerned chiefly with rubidium and strontium. The rubidium measurements comprised eight chemical determinations of the rubidium in rubidium minerals, using separated Rb⁸⁷ as the "spike" material added to the unknown in its original condition. In seven lepidolite

minerals the rubidium content ranged from 1.21 to 3.13 per cent. To obtain age determinations, strontium analyses must be made on the same minerals. The precision of these rubidium analyses is of the order of 3 per cent, but this figure could be brought below 1 per cent without great effort if higher precision were necessary.

The major effort in isotope measurement during the year was the analysis of strontium extracted from the minerals biotite and feldspar, obtained from granite and from celestite, a sedimentary mineral. Samples of these minerals were collected from all parts of the world. This work was carried out in collaboration with Professor L. H. Ahrens, of the Massachusetts Institute of Technology, and two of his students. The initial mineral separation from granites and the rough chemistry for extraction of calcium-strontium concentrates were carried out at the Massachusetts Institute of Technology. Subsequent calciumstrontium separation on the ion-exchange columns, followed by mass spectrometric analyses of the strontium isotopes, was carried out here at the Department. The precision of the strontium isotope analysis is of the order of 0.5 per cent, and the reproducibility on mineral samples which contain sufficient strontium for repeat runs lies within this limit. Approximately 50 micrograms of strontium are required for an analysis with this precision. Six separate analyses on commercial strontium, measured on different days since January 1951, have given the abundances shown in table 1. These are in excellent agreement with those obtained by Nier in 1938 under quite different conditions.

In feldspars, which have a low rubidiumstrontium ratio, the variation in the Sr⁸⁷/ Sr⁸⁸ ratio is from 0.0832 for very ancient minerals to 0.0868 for younger ones or for those with appreciable rubidium content. This ratio in the strontium mineral celestite has been found to vary from 0.0848 to 0.0868. The significance of this small variation in the Sr⁸⁷ abundance in minerals which are low in rubidium lies in the indication it gives that the Sr⁸⁷ content of various strontium minerals, crystallizing out from the mixed world pool of strontium at different times, has not changed appreciably in geologic time. One might have hoped that variation of the Sr⁸⁷ content could have been used as a crude time indicator. This result gives one form of

TABLE 1

Relative abundance of isotopes of commercial strontium

Іѕоторе	RELATIVE ABUNDANCE	
	Our analyses	Nier
88	100 8.50±0.04 11.95±0.03 0.67±0.05	100 8.50 11.94 0.68

evidence regarding the relative geochemical abundance of rubidium and strontium. If, as is now supposed, rubidium and strontium are about equally abundant, then, since the amount of Sr87 formed in the past 1.5 × 109 years should be about 2 per cent of the Rb87 present in the crust, about 0.005 of the total rubidium should have been converted to Sr87. Since Sr87 is about 0.07 of the total strontium, then, assuming equal abundance of rubidium and strontium, this 0.005 rubidium is about 10 per cent of the total Sr⁸⁷ present today. On the other hand, our results indicate that only about 2 per cent of the Sr⁸⁷ in modern sources could have come from the decay of Rb⁸⁷. This means either that a change of a factor of 5 in the present assumption regarding the relative geochemical abundance of rubidium and strontium in crustal material must be made, or that some process in which rubidium was lost to the weathering cycle early in the earth's history must be postulated.

In biotites, which have a relatively high rubidium-strontium ratio, the Sr⁸⁷/Sr⁸⁸ ratio has been found to vary from 0.0838 to 0.2629. The former ratio corresponds to a very young (Tertiary) biotite and the latter to one formed in the Paleozoic era. The variation in Sr⁸⁷ in rubidium minerals (biotite) confirms the prediction made by Ahrens of its suitability for agemeasurement work.

An unexpected and at present unexplained but very interesting part of the work on strontium has been the variation found in the ratio Sr⁸⁶/Sr⁸⁸. Neither of these isotopes has a known radioactive parent. Our work on lepidolites verifies what others have found, that the only product of rubidium decay in ancient minerals is Sr⁸⁷. Yet the variation in the Sr⁸⁶/ Sr⁸⁸ ratio in biotite is from 0.1150 to 0.1210, in feldspar from 0.1165 to 0.1195, and in celestite from 0.1182 to 0.1222. The total range of variation, from 0.1150 to 0.1222, is much greater than has been found in any alkali or alkaline earth element where no known radioactive element is producing the change. The variation found is not related to the mineral age or any other parameter against which it has been tested.

Among other strontium measurements made this year were those correlated with the rubidium spike measurements on lepidolites to obtain age measurements for minerals on which we have done all the analyses, and which are hence independent of any optical spectrochemistry. These results are summarized in table 2. All the ages given in table 2 appear high, but they are independent measurements in which systematic errors of some previous determinations have been eliminated, and at

least they make it a matter of interest to redetermine the decay constants of Rb⁸⁷.

Mr. L. F. Herzog, a student of P. M. Hurley at the Massachusetts Institute of Technology, spent much of the year here using our ion resin column and mass-spectrometer procedures. Our shop also fabricated a mass-spectrometer tube for future use at the Massachusetts Institute of Technology in measuring isotope abundances of calcium, argon, and strontium in separated minerals. Dr. B. F. J. Schon-

performed, not on granite constituents, but on a relatively concentrated ore of the parent chemical element, such that weighable quantities of both parent and daughter could be obtained. It is recognized that at least three advantages should result from the ability to measure ages directly on the minerals separated from granitic material: (1) The granite method will have much wider applicability than the ore method, since granites form a much more abundant and typical crustal constit-

TABLE 2
Ages of Lepidolites

Mineral	Location	$\frac{\text{mg Sr}^{87}}{\text{mg Rb}^{87}} \times 10^2$	Age × 10 ⁻⁶ year (Rb ⁸⁷ decay const = 1.09 × 10 ⁻¹¹ yr ⁻¹)
Lepidolite no. 1 Lepidolite no. 3 Lepidolite no. 4 Lepidolite no. 6 Rb muscovite no. 7	Manitoba, Can. Southern Rhodesia California, U. S. A.	2.26 3.80 3.60 0.170 2.60	2100 3500 3300 157 2400

land was a guest of the laboratory in April and May 1952, in connection with his program for radioactive age determinations and related geochemistry in South Africa.

Granite Ages by Uranium, Thorium, and Lead Ratios

The uranium and lead work on the granite from the Essonville-Tory Hill area of the Canadian shield has progressed during this year to the point where the results are being prepared for publication. The project has served to test the feasibility of making age measurements on granitic rocks on the basis of the different mineral constituents present, with gratifying results.

All previous age determinations based on the decay of uranium and thorium to leads of different atomic weights have been

uent than do uranium and thorium ores. (2) There is the possibility with granites of determining other completely independent ages besides the lead age. Examples are rubidium-strontium and potassiumargon ages. (3) It is reasonable to expect that the discrepancies found in many past age determinations on ore materials will disappear when granitic minerals are utilized, because the crystal structure of minerals is better preserved. The large energies accompanying radioactive decay processes are sufficient to rupture the crystal bonds in a lattice. The greater the amount of radioactivity present, the greater will be this lattice destruction, with resulting exposure to chemical and physical processes capable of influencing the age result obtained. The concentration of uranium and thorium in even the most radioactive granitic minerals is about one

hundred times lower than that in an average ore of these elements.

The average granite is 3 to 4 parts per million uranium, 10 to 15 parts per million thorium, and about 20 parts per million lead. Precise quantitative determination of these elements in such dilute concentrations has been difficult, if not impossible, in the past, and for this reason this method of age determination has not previously been applied to granites. The isotopic dilution method of analysis in conjunction with the mass spectrometer has met the requirements of accuracy for the quantitative determination of all three elements on the microgram scale. It has also been necessary to use surface ionization techniques in the mass spectrometric phase of the work, in order to obtain the isotopic composition of microgram amounts of the elements. Conventional gas sources require milligram amounts, which are difficult to isolate from granitic materials without introducing substantial contamination.

The variety of skills required for this study made collaboration desirable. The minerals of the granite selected for study were separated by Dr. E. S. Larsen at the U. S. Geological Survey. Dr. Claire Patterson performed the lead analyses at the University of Chicago, and uranium analyses have been made here and at the University of Chicago. Dr. M. G. Inghram has supervised all phases of the mass spectrometric work at Argonne National Laboratory.

The results obtained on the zircon mineral of the granite have been most encouraging, and at this point would indicate that accurate measurements can be obtained from granitic minerals. The three ages obtained so far are as follows (in millions of years): U²³⁸/Pb²⁰⁶, 1020±25; U²³⁵/Pb²⁰⁷, 1030±25; Pb²⁰⁷/Pb²⁰⁶, 1060±25. These ages are in good agreement with each other and with the acknowledged

geologic age of the Essonville-Tory Hill area.

Another interesting point has been brought out by this work. It has previously been known that up to about 30 per cent of the radioactivity of a powdered granite can be leached away by the action of dilute acid, which should have no appreciable effect on the silicate minerals present. A uranium-lead study of this nature has been made on the present granite. The results indicate that uranium and lead exist in the acid-soluble fraction in an entirely different ratio to each other from that in the remaining acid-insoluble ("crystal") portion of the rock. The lead has, moreover, a markedly different isotopic composition in the acid-soluble phase from that in the remaining rock.

This type of information may have important bearing on our present theories concerning the isotopic composition of lead in the crust of the earth over past time. Our knowledge to date has been obtained from studies of lead ores of various assumed ages. It is the general belief that these ores are derived from the dilute concentrations of lead existing generally in crustal rock masses. Since the granite studied shows the presence of two distinct types of lead, differing greatly in chemical availability and in isotopic composition, it would appear that lead ore data must be interpreted with considerable reservations.

A thorium carrier for isotopic dilution work has recently been prepared at this laboratory by isolating the thorium from a large sample of pitchblende from the Colorado Plateau (furnished by L. R. Stieff and T. W. Stern, of the U. S. Geological Survey). Natural thorium consists solely of one isotope, Th²³². In the uranium decay chain one part of the daughter isotope Th²³⁰ occurs for each 200,000 parts of uranium. The separation of uranium from thorium in the natural processes pro-

ducing the ore was so complete that it was possible to isolate several hundred micrograms of a thorium which was 80 per cent Th²³⁰. This "spike" was prepared for use in analyzing the zircon of Essonville–Tory Hill granite for thorium and obtaining still another age measurement on it.

The thorium work is of some chemical as well as geological interest, since the detection limit for thorium using the Th²³⁰ carrier will be about o.or microgram of Th²³², which is about one hundred times lower than the detection limit for the best methods in use at present for the detection of that isotope.

Lead-uranium work on a second granite, the Uncompandere, from the Colorado Plateau, has been nearly completed over the past year, although the data are not yet complete enough to state the age of the rock or the isotopic composition of its lead. This work will be of value in providing further evidence on the reliability of age determinations made from granitic minerals, and, coupled with the data from the Canadian granite, will provide the first insight, other than that based on ore data, into the problem of the variation in isotopic composition of crustal lead over geologic time.

Activities planned for the coming year include the necessary thorium determinations on the minerals from the Canadian granite, completion of work on the Uncompander granite, and pursuit of uranium-lead studies on certain basic volcanic rocks, for the purpose of comparing the lead data from these with the data from granites and ores.

It is then planned to synchronize the lead-uranium program with the rubidiumstrontium program and work on the granites dated by the latter method. This will provide some idea of the relative value of the two dating techniques, and the combined information will be used still further to look into the reliability of the potassiumargon method. The latter procedure should be the one easiest and quickest to apply if it can be shown to yield trustworthy dates for Pre-Cambrian rocks.

Magnetization of Rocks

J. W. GRAHAM

In the report for last year mention was made of the problem of evaluating the significance of observations that had been made on flat-lying undeformed Paleozoic sediments. These samples, unlike those from the folded Appalachian mountain sections, have magnetizations distributed in a pattern closely related to the present compass directions, a fact which suggests that the earth's magnetic field has been substantially constant throughout much of geologic time. In order that such a hypothesis could be considered to be proved by the magnetic measurements on rocks, it would be necessary to show that the magnetism measured in the selected rocks today is truly stable and has not simply followed possible changes in direction of the earth's field.

The question of the magnetic stability of ancient sediments was investigated in detail this year by extensive observations of the directions of magnetization of large pebbles embedded in ancient conglomerates, the expectation being that stable magnetizations should be directed much at random, in keeping with the random positions of the pebbles. The results of this work were surprising and at variance with our earlier observations in many parts of the United States, which had demonstrated that many rocks have truly stable magnetizations. At a number of localities in New York state and Vermont, however, it was found this year that the pebbles in the conglomerate are all magnetized in the

same direction and that this direction departs significantly, though only moderately (about 10°), from the direction the earth's field has today. Quite clearly, these magnetizations were brought to a more or less uniform direction when the orientation of the earth's magnetic field was different from what it is at present. Because the direction and angle of departure from the earth's field today are not the same for these conglomerates in all localities examined, we must conclude that the earth's field "wanders," at a minimum, within the limits indicated by these conglomerates. Furthermore, to the extent that the behavior of these early Paleozoic conglomerate pebbles is representative of the behavior of the flat beds, we must conclude that our previous measurements in the undeformed Paleozoic sediments do not give us a completely valid picture of previous directions of the earth's magnetic field.

Our most convincing example of stable polarizations, observed in folded sandstones, was reported in Year Book No. 49 (1949-1950). These rocks are of interest not only because of their remarkable stability, but because their sense of magnetization is almost the opposite of the earth's present field. We have suggested that this inverse magnetization records incorrectly the sense of the field that prevailed when the rocks were formed, and that by some obscure mechanism the rocks have acquired a polarization the opposite of the impressed field. In a private communication this possibility was pointed out to Professor L. Néel at the Institut Fourier, Université de Grenoble, France. He has considered the notion theoretically, and has suggested a number of mechanisms by which such inverse magnetizations could arise when the ferromagnetic constituent consists of two different phases that interact magnetically. Magnetization experiments on these stable "inverse" rocks demonstrate a behavior that is not at all characteristic of a simple single-phase system. For these reasons, we continue to doubt whether these inverse polarizations demonstrate a reversal of the earth's field, although as yet we have no direct proof of a mechanism in these rocks for self reversal of the magnetization.

Because we have been able to demonstrate that in many cases the residual magnetization of Paleozoic rocks gives an unreliable indication of past directions of the earth's magnetic field, we have turned our attention in addition to a relatively unexplored technique for inferring some of the early history of that field (see Year Book No. 49, 1949-1950, for earlier details). This method is based on the idea that at the time the sediments were deposited, the tiny grains of ferromagnetic minerals may have become oriented with their longest dimensions parallel to the earth's field, thus giving the sediments a larger magnetic susceptibility along this axis. Basically this technique is attractive, for it does not depend on so delicate a property as a direction of magnetization, but depends instead on the ability of small particles to retain their shape and orientation and their magnetizability. The shape and orientation of these grains can be determined on a statistical basis by measuring the ease of magnetization (magnetic susceptibility) of the rock sample along different directions. During the report year, a useful instrument for such measurements was developed. It is sensitive to as little as 10-8 cgs/cc of susceptibility, and the ellipsoid, very little different from a sphere, which describes statistically the shape orientation of the grains can be measured directly in a few minutes.

Observations made during the year suggest that the direction of maximum susceptibility (the greatest elongation of the

grains) is controlled both by the earth's magnetic field and by mechanical processes that affect the sediments during and after deposition. For example, the direction of maximum susceptibility of some flat-lying Silurian and Devonian limestones from New York is horizontal and directed north-south, and the minimum susceptibility is vertical. The same result was obtained from samplings of a dried mud puddle on the laboratory grounds. We suspect that in both cases the drying down and compacting of the sediments may have caused the elongated grains to rotate toward a horizontal position, so that the true inclination of the earth's field is not indicated; the declination may be retained. Further studies are in progress to determine whether this technique will have real value to geologists in describing the deformation of rocks, and in deriving the history of the earth's magnetic field.

UPPER ATMOSPHERE RESEARCH

H. W. Wells

A major effort this year has been directed toward the elimination of certain gaps in our knowledge of basic properties of the upper atmosphere. Experimental observations have continued in the general field of atmospheric "dynamics" with special attention to the solar eclipse of September 1, 1951. Instrumental development has continued with the construction of units to record precise height fluctuations and amplitude changes of ionospheric echoes. The installation of a radio interferometer for solar observations is nearing completion at Derwood. Much progress has been made toward the instrumentation necessary to measure monochromatic radiofrequency radiation from the Galaxy. Studies of the extremely high atmosphere from F-region scatter have been continued. Favorable theoretical analyses led to the initiation of an experiment to test the effectiveness of this type of long-range propagation at ultra high frequencies. Active liaison and co-operation have been maintained with government agencies and technical organizations.

June 1952 finds the three-station experiment over short base lines in active operation. As in earlier experiments, the Derwood installation is used as the base station. The trailers with complete ionospheric sounding instruments are located approximately 20 km west and 20 km north of the Derwood station. The westerly unit is located on a farm south of Poolesville, Maryland, and the northerly unit is installed on the grounds of the high school at Damascus, Maryland.

Although a complete factual report cannot be prepared until the observations have been completed and analyzed, some of the early results available at this time are of considerable interest. In particular, the recordings of several sporadic E clouds have indicated apparent velocities of only 2 to 3 km per minute. This is approximately 30 to 50 m per second. Dimensions of two cloud patches have been estimated at 40 and 50 km, and the apparent motions have been to the southeast and northwest. The new close spacing of the three observing stations appears to remove any uncertainty in the identification of the same ionospheric events at all the stations.

The present experiment has evolved from the three-station operations between Derwood, Charlottesville, Virginia, and Morgantown, West Virginia; the operations between Derwood, Charlottesville, and Chincoteague, Virginia; and the line experiment between Derwood, Charlottesville, and a point one-third of the distance to Charlottesville, which were discussed in last year's report. The operations over base lines of 100 to 150 miles showed the occurrence of numerous local disturbances

at each station which could not be positively identified at the other stations without laborious statistical operations. The line experiment of 1951, which coincided with a period of extended magnetic disturbance, showed occasional large differences in ionospheric properties, especially sporadic E, between Derwood and a station approximately 50 km distant.

The eclipse operation of September 1, 1951 was concluded with a high degree of success. The results have been analyzed in considerable detail, and one report on the principal events and interpretations associated with the eclipse has been completed. The results show absence of significant ion production at any station until more than one-third of the sun was exposed during the recovery phase of eclipse. Just after sunrise, small increases in F2 ionization were noted at two stations where the sun was 46 and 54 per cent eclipsed, respectively. See figures 1 and 2.

At the third station, however, where the sun was 66 per cent eclipsed at sunrise, no increase was noted until the recovery phase, as remarked earlier. Computed values of ion density during the eclipse period, which were based on numerical integration methods, showed excellent agreement with observations until some 15 minutes after maximum phase, at which time the observed rate of increase of ionization was nearly twice as great as could be accounted for on the basis of normal ionizing processes. It may be inferred that other factors, such as transport, were actively contributing to the over-all ionization during the recovery phase of eclipse.

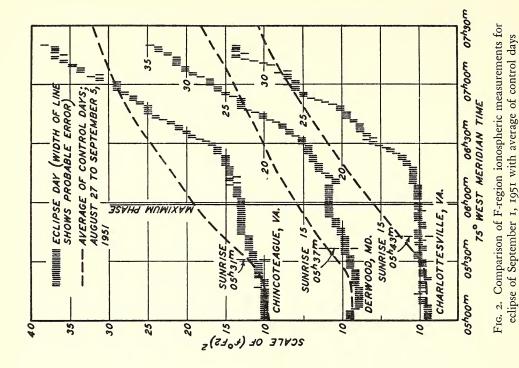
The statistical methods developed by Briggs, Phillips, and Shinn have been applied to the determination of the average apparent drift velocity of F-region irregularities. The values fall mostly between 600 and 1200 km/hr (160 to 320 m/sec). Estimates of the size of these F-region disturb-

ances vary between 100 and 300 km. No preferred directional pattern has been indicated. When results on the F-region reflections obtained from the short-base operation are available, it will be possible to assess the significance of average values by correlation analyses.

The records made in connection with the eclipse are being analyzed for vertical as well as horizontal components of traveling disturbances. Scalings of virtual heights were obtained from minimum F-region height and at two frequencies, 5 and 6 megacycles. Layer tilts or slopes between 10° and 15° were apparent on several occasions.

The dual-channel recorder was developed to a point where intermittent records of heights and amplitudes could be obtained at a fixed frequency. The instrument was subsequently dismantled and shipped to L. V. Berkner at the Brookhaven Laboratory. The relatively simple panels of a new type of auxiliary pen-andink recorder for heights and amplitudes (M. H. MacKenzie) were used to obtain characteristic data for different hours and frequencies. The results of some analyses showing fine structure and periodicities of oscillations were presented at a regular staff meeting.

A considerable amount of attention has been given to methods of reducing the man-hours involved in the reduction and analysis of observations. Three independent methods of recording have been tried. All these use slowly moving film instead of the intermittent-frame methods of earlier applications. One method using side-by-side recordings on slowly moving film was tested during a month of observations at Derwood. Analyses of F-region perturbations in critical frequency show a pronounced interdependence between the period and the magnitude. The fluctuations of short period are of small magnitude,



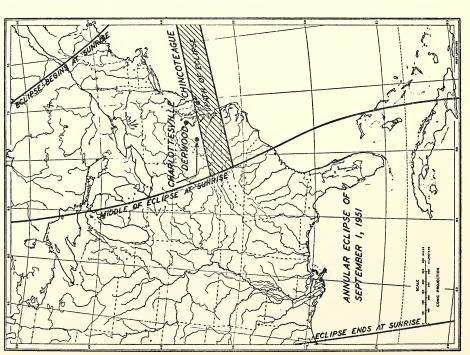


Fig. 1. Observing network for annular eclipse of September 1, 1951

whereas those of longer duration are of greater amplitude. A mass plot shows a "preferred" period of 10 to 15 minutes.

Other projects which are in a stage of advanced instrumentation are in the fields of radio astronomy and long-distance scatter propagation. Tests and adjustments of a radio interferometer with element spacings of 50 to 100 wave lengths are under way at Derwood. A joint project with the section on Physics of the Earth's Crust has been the assembling of the electronic instrumentation necessary for the study of monochromatic radiofrequency radiation in the Galaxy. The initial effort is directed toward extending the Ewen-Purcell measurements of the 1420-megacycle radiation of the hyperfine structure of atomic hydrogen, in our Galaxy and the Andromeda nebula.

Our interest in properties of the extremely high atmosphere was discussed in last year's report. The possibility of an association between F scatter, as established

by analyses of ionospheric records at Huancayo, Peru, and the occasional reports of very long-distance amateur contacts at 50 megacycles was discussed with Professor H. G. Booker, of Cornell, while he was here as a guest investigator. Preliminary theoretical work looked promising, and an experiment was set up to test the idea.

Since April 1952, Dr. John W. Findlay, of the Cavendish Laboratory, Cambridge, England, has been vigorously participating in the activities of this group. His primary interest at this time is in extending our knowledge of traveling disturbances in the ionosphere.

Interesting developments of considerable practical importance in the field of long-distance communication, an outgrowth of many years of study of scattering sources or patchiness in the ionosphere, were announced during the year in a paper in the *Physical Review* with L. V. Berkner, former member of the Department, as one of the joint authors.

STATISTICAL AND OBSERVATORY GEOPHYSICS

THE MAIN MAGNETIC DIPOLE OF THE EARTH

E. H. VESTINE

Astronomers have found from accurate time determinations, based on observations of the stars, the moon, and Mercury, that the earth does not rotate at a uniform rate. After suitable corrections based on the interactions of astronomical bodies and on the slow deceleration of the earth due to tidal action in shallow ocean areas, there yet remains a considerable and puzzling fluctuation in the rate of the earth's rotation during time intervals as short as fifty years.

It now appears likely that an explanation of this outstanding riddle has been discovered; it is related to slow changes in the magnetic field of the earth. From a

study of the time fluctuation in the rate of westward drift of the world-wide geomagnetic field during the period 1885-1945, it has been inferred (see Year Book No. 48, 1948-1949) that there is a corresponding time fluctuation in motion westward of the outer portion of the earth's liquid core. The changes in motion of the outer core are about 25 times as rapid as those found from astronomy for the earth's surface, and in the opposite sense. The irregular motion of heavy liquid in the earth's core, shown by the magnetic changes, is precisely what is required to explain the fluctuation in the rate of the earth's rotation on the basis of conservation of angular momentum.

This deduction is checked in a remarkable way. The geomagnetic data show that

the westward fluctuation in motion of the earth's outer core is accompanied by a fluctuating motion of the outer core about an axis perpendicular to the earth's axis of rotation. This transverse supply of angular momentum, with the same moment of inertia required for explaining the fluctuation in rate of rotation, is adequate to explain the year-to-year variations of latitude observed by astronomers. Preliminary calculations give a shift of the north pole, relative to the axis of rotation, of about 12 feet from 1910 to 1940, in good accord with some studies by astronomers. Hence, two major geophysical puzzles are explained by the same mechanism. Other existing experimental data on the earth and atmosphere show that only in the core does there exist the capacity for the rapid and large changes in angular momentum required.

A similar study is being pursued with a view to obtaining a better understanding of the known smaller seasonal fluctuation of the earth's rate of rotation. It is known from previous work that a part of this is explained by changes in the angular momentum of zonal air motion in the lower atmosphere. If correlated zonal air motions in the upper atmosphere also are rapid enough and extensive enough, there may also exist a correlation between magnetic changes originating above the earth's surface and the earth's rate of rotation. Calculations were made of the electromagnetic and gravitational couples due to solar streams of emitted particles, and to the ring current in the equatorial plane at several earth radii postulated by the Chapman-Ferraro theory. These show the supply of angular momentum from such sources to be too small by several orders of magnitude to contribute sensibly to the observed seasonal fluctuation in the earth's rate of rotation. Hence, it is important to

show whether or not a dynamic connection with short-term geomagnetic changes exists. So far, the correlations have given no decisive results, but they are continuing in co-operation with the U. S. Naval Observatory.

A theoretical study, motivated by eclipse results of F2-region ionization, was made of the origin and evolution of the lunar atmosphere. It was shown that although the accretion and escape of gases from geological and solar sources may lead to an atmosphere including carbon dioxide and sulfur dioxide, the pressure at the lunar surface probably does not exceed that at the F region of the earth's atmosphere, or about 10⁻¹⁰ atmosphere. This amount of atmosphere is further reduced by photodissociation.

COSMIC-RAY RESEARCH

CORRELATION OF COSMIC-RAY IONIZATION
AT HIGH ALTITUDES AND AT SEA LEVEL,
AND NEUTRON INTENSITIES ON
MOUNTAIN TOPS

S. E. Forbush

In collaboration with Dr. H. V. Neher, California Institute of Technology, the results from his extensive series of ionization measurements from high-altitude balloon flights have been correlated with those from Carnegie Institution of Washington meters at Cheltenham and Huancayo and also with the results of neutron measurements published by J. Simpson (*Phys. Rev.*, vol. 85, p. 366, 1952).

The balloon flights were made over a period of several weeks in the summer of 1951 at an altitude of about 70,000 feet over Bismarck, North Dakota. The neutron data were obtained at Climax, Colorado. The variations in ionization at Bismarck over the period involved correlated well with those from CIW meters at Chelten-

ham and Huancayo and with the variations in neutron intensity at Climax. The magnitude of the variation in ionization, about 12 per cent, at 70,000 feet was roughly seven times that at Cheltenham, about six times that at Huancayo, and about twice the magnitude of the variation in neutron intensity at Climax.

These variations have the characteristics of the 27-day quasi-periodic changes in cosmic-ray intensity which occur both at Cheltenham and at Huancayo, and are correlated (negatively) with the 27-day changes in magnetic activity; thus they are quite distinct from the solar-flare effect, which has never been observed at Huancayo. Whatever mechanism brings about the former, it involves primaries of much higher energy than are involved in the solar-flare effect. Since higher-energy primaries are less effective in producing nucleons, they also account for the fact that the percentage variation in neutron intensity at Climax (altitude 11,000 feet) was only about three times that in ionization at Cheltenham for the variations under discussion, whereas during the solar-flare effect of November 19, 1949,

the percentage increase in neutron intensity observed near sea level at Manchester, England, was from 15 to 60 times the percentage change in ionization at Cheltenham.

Neutron meters. One neutron ionization meter has been maintained in practically continuous operation since October 1951 at the Derwood Experimental Station. The barometric coefficient derived from several months' data agrees very well with that derived from neutroncounter data obtained by the group at Cornell University. This provides reasonable assurance that these two types of instrument should give similar results for variations in neutron intensity due to solar flares or other causes. A second neutron meter is now being assembled for tests at Derwood and for comparison with the first, prior to installation at Climax or Jasper Park.

Old cosmic-ray program. Operation of Carnegie Institution of Washington Compton-Bennett meters was continued at Godhavn (Greenland), Cheltenham (Maryland), Huancayo (Peru), and Christchurch (New Zealand).

LABORATORY PHYSICS

NUCLEAR PHYSICS

N. P. HEYDENBURG, D. B. COWIE

The investigation of alpha-particle-induced nuclear disintegrations and alpha-particle scattering, for energies below 7 Mev, has been made with natural radio-active sources. The accuracy and scope of these experiments have been limited by the low intensity of these sources. The recent development of radiofrequency-excited ion sources has made good currents of helium ions available from electrostatic generators. Early in the report year, ion sources of this type were installed on the

Department's one-million-volt and three-million-volt electrostatic generators. A program of study of the disintegrations produced by alpha particles (helium nuclei) and of alpha-alpha scattering was then initiated. An extended series of measurements has been completed.

Several aspects of alpha-alpha scattering are of interest for alpha-particle energies below 7 Mev. Simple theoretical considerations require that with energies of the order of several hundred kev the interference effect of scattered and recoil alphas should become apparent. Maxima and minima should occur in the scattering

cross section as a function of the angle of scattering. This behavior occurs only when the particles scattered are identical, and the interference effect depends on the nuclear spin and statistical nature of the alpha particles involved.

At some energy above this region, the scattering is expected to deviate from the scattering law based on the Coulomb repulsion of the two particles. This will occur when the nuclear force becomes comparable to the Coulomb force. A detailed study of the scattering in this energy region should yield information about the interaction potential between alpha particles. The scattering will also be affected by the presence of energy levels in the nucleus beryllium of mass eight, which is the compound nucleus for the interaction of two alphas. The Be8 nucleus is known to be unstable by about 80 kev to the disintegration into two alpha particles. The first excited level of Be8 is 3 Mev above the ground level. The alpha-particle model of nuclear structure, which is based on the idea that the stable alpha-particle structure persists as a grouping in more complex nuclei, requires that this level should have angular momentum J=2; that is, it must be a D level. The analysis of alpha-scattering data obtained with natural sources is not sufficiently precise to decide whether I=0 or 2 for this level.

The angular dependence of alpha-alpha scattering has been measured for energies from 150 kev to 3 Mev. The results at the low energies are in good agreement with Mott's quantum theoretical calculations, in which the interference effect of the scattered and recoil particles is taken into account, assuming that the alpha particles obey Bose-Einstein statistics and have zero spin.

Above energies of 400 kev the angular distribution of scattered alphas did not agree with curves calculated from the Coulomb interaction alone. For energies

up to 2.5 Mev the anomalous scattering effect could be accounted for by assuming only zero angular momentum, or S-wave, nuclear interaction between the alphas. The S-wave phase angle varied from 180° near 400 kev to 135° at 2.5 Mev. This behavior is in contrast with that found in proton-proton scattering, where the phase angle increases from o° with increasing energy. If the ground level of Be8 is an S level (zero angular momentum), then the phase angle should increase from o° below the level energy to 180° above this energy. Since this level is 80 key above the rest energy of two alphas (160 kev in the laboratory co-ordinate system), any change in phase angle above 160 kev should start from 180°. Observations on the scattering cross section at 45° scattering angle, in the region of energies near 160 key, failed to show this 180° change in phase. From theoretical considerations it is probable that the ground level of Be8 is very narrow and its effect on the scattering would not be seen because of energy spread of the alphaparticle beam, which is roughly 500 ev at this energy setting of our equipment.

The 3-Mev excited level of Be⁸ has considerable breadth, so that its effect on the scattering may show up well below the energy of the level. If it is a D level, then a D-wave phase angle should be required to account for the scattering distribution. No appreciable amount of D phase angle was required for the scattering data at 3 Mev (1.5 Mev in the center-of-mass coordinate system). It may be possible to extend the energy range of these measurements by use of the singly charged 8-Mev helium beam from the cyclotron to cover this energy region of the 3-Mev Be⁸ level completely.

Two nuclear reactions produced by alpha-particle bombardment have so far been studied. This work was done in collaboration with Dr. F. L. Talbott, of Catholic University. The yield of gamma rays

and neutrons from beryllium bombarded by alpha particles was first investigated. The reaction can be written as follows: Be 9 + alpha \rightarrow C 13 \rightarrow C 12 + neutron + gamma. The yield of both gamma rays and neutrons increased sharply above 1.5 Mev to a resonant peak at 1.92 Mev. A smaller resonance occurred at 2.60 Mev. These resonances correspond to energy levels in the compound nucleus C 13 at 11.95 Mev and 12.43 Mev.

In the second reaction studied, B^{10} + alpha $\rightarrow N^{14} \rightarrow C^{13}$ + proton + gamma ray, the gamma-ray yield was determined as a function of the bombarding energy. Thin boron targets containing 96 per cent B^{10} were used. Five sharp resonances, having half widths above 25 keV, and two broad peaks were observed. These resonance peaks correspond to energy levels in N^{14} at 12.77, 12.87, 12.90, 13.01, and 13.22 MeV. The broad peaks appear to be due to a number of unresolved overlapping levels.

The 7.5-Mev proton beam from the cyclotron was used to investigate inelastically scattered protons from twelve light nuclei, from beryllium through neon. A typical reaction of this type is the following: $Be^9 + proton \rightarrow *Be^9 + proton$. The beryllium nucleus is left in an excited level, and the inelastically scattered proton has lower energy by the amount of excitation. The nucleus returns to its ground level with the emission of a gamma ray. Since this process occurs only for discrete energy levels of the nucleus, it is a very useful method for locating energy levels. Eighteen levels were observed in all. Most of these agreed in energy with levels determined from other nuclear reactions. A new level was observed at 3.83 Mev in F¹⁹.

BIOPHYSICS

The advent of artificially radioactive isotopes of ordinary chemical elements, produced by deuteron or neutron bombard-

ment, and the simultaneous rapid development of mass spectrographs and the means for separation of stable isotopes led to the clear realization that the use of isotopes as tracers in following the metabolic pathways of normal chemical elements and compounds in living tissue would be a fruitful field of effort for many years. The need for very special knowledge and interests among the investigators who might work in this field was obvious. The Institution undertook in 1938 to provide suitably large equipment and facilities adjacent and supplementary to those used in the nuclear physics activities of this Department, which had been vigorously in progress for many years, in order to enable the biological research laboratories in the Washington area to initiate work with isotope tracers. The postwar development of isotope manufacture and distribution by the Atomic Energy Commission has removed the load of producing isotopes with the Department's 60-inch cyclotron for other research groups. The physicists of the Department's nuclear physics group, however, have not abandoned collaboration with biology; instead, they have largely directed their interests toward studies of biological processes using isotopes. The physicists here view these challenging problems in living matter in much the same way as a physicist examines process and order in the organization of matter and energy to form a crystal or a semiconductor or some other conventional physical aggregation.

Dynamics of Synthesis

P. H. ABELSON, E. T. BOLTON, R. J. BRITTEN, D. B. COWIE, R. B. ROBERTS; S. LÖVTRUP

During the past year this group has continued to focus its attention on problems related to protein and nucleic acid synthesis. Protein synthesis has been examined from several approaches, including studies

of (1) the synthesis of the constituent amino acids, (2) the formation and utilization of peptides, (3) the formation of adaptive enzymes, and (4) amino acid sequence in the finished proteins. Studies have also been conducted on the synthesis of the constituents of nucleic acids and the relation of nucleic acid to protein synthesis. These individual studies form a closely related whole where each reinforces and clarifies the others. This area appears to be most promising at present for studies directed toward increasing our understanding of the processes by which a single cell can organize the material and energy from its surroundings for reproduction of itself. Without protein and nucleic acid no life exists. The duplication of these large molecules seems to be the key to the duplication of the cell as a whole.

Special techniques in physics and chemistry developed or refined in recent years have greatly facilitated the study of these problems. Ion-exchange resins permit the rapid separation of various types of cellular components. Paper chromatography and paper electrophoresis permit the separation and measurement, in micro quantities, of very similar biological products. Radioactive tracers make it possible to follow individual atoms through the complex chemical changes which occur in the synthetic process. By combining these methods it is possible to observe the basic mechanisms of biosynthesis as they occur in normal growing cells. For example, with radioactive sulfur the incorporation of inorganic sulfur into the tripeptide glutathione and finally into the proteins can be observed. Or, with radioactive carbon the incorporation of carbon dioxide into the nucleic acids and proteins can be followed. The exact mechanisms concerned can be studied by using tracer molecules suspected of being metabolic intermediates or

by using unlabeled intermediates to compete with labeled compounds.

Escherichia coli has been used as the principal test organism because of its convenience and rapid growth, and because many of the basic biochemical processes seem to be similar throughout all living systems. For example, the same pathways of carbon dioxide fixation are found in E. coli as in mammalian tissue, yeast, and mold. Going into greater detail, the synthesis of arginine by way of citrulline and ornithine, first suggested by experiments with liver slices, is the same—atom for atom—in E. coli. In some cases minor differences are noted: the sulfur metabolism of yeast and mold appears to be different from that of E. coli. In general, however, it is possible to uncover processes of widespread biological importance by the study of bacteria.

Amino Acid Studies

Almost all proteins derived from a wide variety of different organisms are built up from combinations of approximately twenty universal amino acid building blocks. Consequently the synthesis of the amino acids themselves may be regarded as the first step in the synthesis of proteins. This step is relatively independent of later stages in protein synthesis, as many cells are unable to synthesize amino acids but have no difficulty in assembling them into proteins.

In last year's report the results of a study of C¹⁴O₂ uptake and utilization by *E. coli* were presented. In these experiments glucose was the sole source of energy and major source of carbon for synthesis. Cells grown in the presence of C¹⁴O₂ used this compound as a building block, and examination of hydrolysates of the bacteria discovered C¹⁴ in eighteen different compounds including seven amino acids: as-

partic acid, glutamic acid, arginine, isoleucine, lysine, proline, and threonine. Recent studies have shown that other amino acids including methionine are also labeled to a slight extent. The question arose whether the patterns of incorporation were related to a specific sequence of reactions starting from glucose, or whether patterns of C14O2 incorporation might be largely independent of energy and carbon sources. Escherichia coli which have been grown with glucose as an energy source can be adapted to use other energy sources if glucose is not present. Thus, after two hours' incubation in the presence of glycerol, the cells resumed growth using this substance as a carbon and energy source. In a similar manner the bacteria were adapted to lactate, pyruvate, succinate, alanine, serine, and α-ketoglutarate. The adapted bacteria were subsequently grown in the presence of the appropriate energy source and C14O2.

After suitable extraction procedures to free the cells of nonprotein constituents, the proteins were hydrolyzed and the appropriate chromatographic separations and radioautographs made. These revealed that in general the pattern of C¹⁴O₂ fixation into purine, pyrimidines, and amino acids was strikingly similar in the various cultures, independent of energy source. Thus in each case C¹⁴ was found in the same amino acids when glucose was the principal energy and carbon source and when, for example, glycerol or pyruvate was the energy and carbon source.

In most cases the relative amounts of $C^{14}O_2$ fixed in the various amino acids were the same. There were a few exceptions to these findings. Thus, when succinate and α -ketoglutarate were the energy sources, there was a marked alteration in the quantitative ratios of labeling of the amino acids, although the qualitative pattern was preserved. With alanine as the

energy source, the usual pattern was present and an additional amino acid, glycine, was found to contain substantial amounts of C¹⁴.

Further information concerning the pathways of amino acids can be gained by using various other compounds labeled with C¹⁴ as either sole or supplemental carbon sources. Glucose labeled in the C₁ position was used in a series of experiments. The C₁ carbon is found in aspartic and glutamic acid and in the family of amino acids which were shown to be derived from them by the C¹⁴O₂ work (arginine, lysine, proline, threonine, and isoleucine). In addition, the C₁ carbon appeared prominently in alanine, tyrosine, and valine.

More detailed information concerning the pathways of amino acid synthesis and interconversions can be gained by utilizing C14-labeled amino acids themselves as supplements to the medium. Experiments have been made to date employing aspartic acid, glutamic acid, glycine, serine, alanine, threonine, valine, proline, leucine, and isoleucine. These studies, which are still in progress, have shown that aspartic acid, glutamic acid, serine, alanine, and threonine are used in a variety of synthetic pathways by E. coli. Glycine, proline, leucine, and isoleucine are used by the cells but are not appreciably converted into other amino acids.

As most of these amino acids were not available commercially, it was necessary to prepare them in the laboratory. Algae that can use carbon dioxide as a sole carbon source, *Chlorella pyrenoidosa*, were grown in the presence of C¹⁴O₂. The entire protoplasm of the cells was uniformly labeled with the tracer and provided a source of many compounds. After the growth period, the cells were harvested and the proteins freed of nonprotein constituents. The radioactive protein was hydrolyzed with

6n HCl and the individual amino acids were separated and purified by procedures including anion- and cation-exchange resins and paper chromatography.

The work on the sulfur-containing amino acids cystine and methionine, which was reported last year, has been extended by comparing the sulfur metabolism of a yeast (Torulopsis utilis) and a mold (Neurospora crassa) with that previously observed in E. coli. In E. coli, cystine sulfur is converted to methionine sulfur presumably by way of homocystine, but there is no evidence from our studies that cystathionine is an intermediate. Furthermore, the reverse reaction methionine to cystine does not readily occur. Using the same technique it has been demonstrated that cystathionine plays a part in the sulfur metabolism of the yeast and mold. Also, methionine sulfur is rapidly converted to cystine sulfur in both. Externally supplied cysteine is not utilized by E. coli, but is preferred to cystine in the yeast and mold. Further studies of the permeability of these organisms must be made before a final interpretation of these results can be given.

Glutathione

As a second step in protein synthesis, the amino acids may be linked together in groups of two or three, and these subassemblies (peptides) may subsequently be used in the synthesis of the complete protein. This hypothesis is attractive from several points of view, but to date there is little evidence for it. E. F. Gale, in England, has observed the formation of peptides when protein synthesis was blocked by chloramphenicol, but similar observations have not been made on normal growing cells. Also, one hexapeptide has been split off a protein and shown to be formed separately from the protein. Further evidence is required, however, to establish the hypothesis that peptide formation is a general or important step in protein synthesis.

Consequently, we were pleased to identify one distinctive fraction obtained from *E. coli* as glutathione, a tripeptide containing glutamic acid, cysteine, and glycine, which is a compound of widespread biological occurrence. Since this molecule contains sulfur, it can be tagged easily with radioactivity and its kinetic behavior observed. This use of radioactive tracers and the competition methods made it possible to establish the source of many of the atoms of the glutathione. As is shown in figure 3, when the cells are grown in a

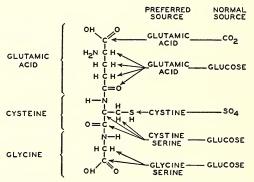


Fig. 3. Pathways for synthesis of glutathione

medium containing glucose and carbon dioxide as carbon sources, all the carbon except the carboxyl carbon of the glutamic acid is derived from glucose. By use of tagged glucose competing with unlabeled amino acids, as well as random tagged amino acids, it was possible to show preferred sources of carbon, as indicated. Also, the source of the sulfur atom could be traced to sulfate with cystine (but not homocystine or methionine) as a preferred source.

The kinetics of formation of glutathione were studied with radioactive sulfur as an indicator. The tracer was added to a culture of growing cells, and samples were

taken at intervals to measure the rate of incorporation of the tagged atom, originally present as sulfate, into the glutathione and proteins. The rate of synthesis of glutathione is high, roughly 3 per cent per minute, as compared with 1.1 per cent per minute for the cells as a whole. As only 1.1 per cent of the 3 per cent is needed to supply glutathione for new cells, the rest must be lost or utilized to form proteins. A part of it does leak out of the cell into the medium (and is used in turn by other cells, since glutathione is a preferred source of sulfur). The remainder, however, seems to be incorporated into the proteins, but the mechanism cannot be determined by this type of experiment, because only the sulfur atom is observed. The rate of glutathione synthesis and utilization is markedly affected by the presence of methionine. The rate of synthesis drops to one-half when methionine is added to the medium, but the total glutathione content of the cell remains the same. Consequently, the rate of utilization must be decreased, and this is the first indication that glutathione is implicated in the synthesis of methionine.

The utilization of the sulfur of glutathione was observed more directly by growing the cells in the presence of radioactive sulfate so that both the glutathione and the proteins became labeled. The cells were then washed and allowed to grow again in an unlabeled medium. During this period the radioactive sulfur decreased in the glutathione while the radioactivity of the proteins increased, showing a transfer of sulfur from glutathione to the cysteine and methionine of the proteins. In addition, some of the radioactive sulfur of the glutathione left the cells, partly in the form of glutathione and partly as another peptide. It is not yet possible to demonstrate whether the glutathione is incorporated into the proteins as a unit or whether the molecule contributes individual amino acids or dipeptides. The combination cysteinyl-glycine, however, is abundant in the proteins, a fact which suggests that at least this part of the glutathione is utilized as a unit for protein synthesis.

Several other features of glutathione metabolism have been uncovered which indicate its importance to the cell. If the sulfur source of the medium is inadequate, the glutathione content of the cell drops abruptly, showing that the glutathione may serve as a reserve supply of sulfur. Also the synthesis of glutathione is relatively insensitive to dinitrophenol. This poison interferes with the formation of the energy-rich phosphate bonds which are energy-transferring mechanisms of the cell. Consequently, glutathione may be synthesized by some other energy-transfer mechanism and act as an energy carrier itself.

Adaptive Enzymes

A unique way in which to investigate the synthesis of protein is to study the formation of enzymes during adaptation. Adaptation manifests itself in the production of new protein, an enzyme, which is specifically capable of preparing a new substrate for utilization by the cell. The mechanism of formation of the new enzyme has been investigated, using as a type system the adaptation of E. coli strain B to the sugar lactose. While they are in the process of adapting to lactose, the cells, which previously had utilized glucose, increase the quantities of the lactose-splitting enzyme, lactase, by a factor of a hundred to a thousand. A considerable increase in the production of this particular kind of protein occurs during the lag phase of the growth cycle, in which actual cell division does not take place.

Lactase is produced in response to the

presence of lactose even when cell division is made impossible by eliminating an external source of nitrogen. Since nitrogen is required for the formation of protein, it is evident that cellular constituents supply this need. It is possible to remove some of the nitrogenous compounds of low molecular weight from the cell by simple washing procedures. When this is done, both the rate of synthesis of lactase and the final amount produced are lowered. The rate of synthesis may be increased (for washed cells) by adding traces of an energy source such as glucose or an energydonating compound such as adenosine triphosphate. Once the low-molecular-weight constituents have been removed from the cell, it is possible to increase the total amount of enzyme only by replacing the lost nitrogen with a source such as an amino acid. Radioactive sulfur, which is a convenient indicator of protein synthesis, shows a greater uptake of isotopic sulfate during adaptation in the absence of external nitrogen than is found in the presence of the normal substrate, glucose.

These results demonstrate that new protein is synthesized during adaptation through the utilization of small molecules. They also suggest that lactase is formed in this way rather than by remolding an already existing protein.

Protein Structure

Other clues to the mechanism of protein synthesis may be found in the structure and composition of the proteins themselves. A few studies of the amino acid sequence of purified individual proteins have been made in other laboratories, but these must be repeated on many different proteins before any generalizations can be made. The viewpoint was adopted here that the total proteins of an organism would provide an average of many indi-

vidual proteins and that any patterns of amino acid sequence found in this average might be characteristic of the modes of synthesis employed by the organism.

It was of particular interest to determine whether the order of amino acids in such an average mixture of proteins would be random or whether some of these building blocks would be more likely to be found together. For this purpose a study was made of the amino acids associated with cysteine in the proteins of *E. coli*. The bacteria were grown in the presence of S³⁵O₄ and ordinary methionine. As a result, the cysteine found in the proteins was radioactive. No other amino acid was labeled during this growth. The cells were harvested and nonprotein constituents removed.

The residual proteins were partially hydrolyzed into small fragments. In any partial hydrolysis a mixture is obtained consisting of free amino acids, dipeptides, tripeptides, tetrapeptides, and larger peptides. If treatment is too severe (6n HCl, 3 hours at 100° C), the fragments are principally free amino acids. If treatment is too mild (6n HCl, 3 minutes at 100° C), the peptides obtained are too large. The exact optimum conditions for producing small peptide groups have not been established, but treatment with 6n HCl at 100° C for 30 minutes was found to be satisfactory.

The studies were facilitated by a chemical procedure, oxidation of cysteine to cysteic acid. Peptides containing cysteic acid were readily separated from the complex hydrolysis mixture by a series of steps including electrophoresis and resin chromatography. The cysteic acid peptides were then separated from one another by paper chromatography, and subsequent radioautographs determined the location on the paper of radioactive peptides. These were eluted from the paper and their amino acid content was identified and determined.

The important new result of these experiments was the demonstration that the amino acids found in association with cysteine were by no means a random assortment. Aspartic acid, glutamic acid, and glycine are generally associated with cysteine, whereas alanine, isoleucine, leucine, phenylalanine, proline, tyrosine, and valine rarely are found in dipeptides and tripeptides containing cysteine.

Further studies to determine patterns of amino acids associated with arginine are under way. It will be necessary to extend these studies and to include other organisms before the general significance of such patterns can be evaluated.

Nucleic Acid Synthesis

Last year's report described the utilization of C14O2 in nucleic acid synthesis. During processes of cell growth and division, radioactive carbon of sodium bicarbonate was extracted from the medium by E. coli and incorporated into purines and pyrimidines. These incorporation processes were specifically altered by culturing cells in media containing as carbon substrates the kinds of purines and pyrimidines which are part of the nucleic acid molecule. Although the uptake of radioactivity was specifically suppressed in these experiments, no change occurred in the proportions of the constituent parts of nucleic acid. The pathways for C14O2 incorporation into nucleic acid were also found to be practically independent of those involved in amino acid synthesis.

To test the possibility that these findings were characteristic merely of bacteria grown in glucose, cultures adapted to utilize other kinds of primary energy source were examined. These other substrates were glycerin, lactate, pyruvate, alanine, serine, succinate, and α -ketoglutarate. Examination of purines and pyrimidines

produced by the various cultures showed that in all instances adenine, guamine, cydidylic acid, and uridylic acid contained large amounts of isotopic tracer. Indeed, examination of radioautographs of the various paper chromatograms showed that all the patterns were virtually identical with those obtained from glucose-grown cultures.

In order to broaden the basis of our understanding of nucleic acid synthesis, studies with radioactive metabolites other than carbon dioxide have been initiated. Amino acids uniformly labeled with C14 were prepared using the green alga Chlorella, and supplied to E. coli. It has been found that alanine contributes only traces of radioactivity to nucleic acid, whereas glycine, serine, threonine, and glutamic acid contribute appreciable amounts. Glycine, serine, and threonine labeled only the purines, whereas glutamic acid labeled both purines and pyrimidines, the latter to a higher degree. This observation is a direct indication of the possible source of the non-carbon dioxide portion of the pyrimidines.

Nucleic acid synthesis has also been studied in the amphibia Rana pipiens and Amblystoma punctatum. The techniques of embryology have been coupled with the analytical methods used in the bacterial studies to yield new information about the formation of nucleic acid during embryogenesis.

Sodium bicarbonate labeled with C¹⁴, when supplied to developing embryos, labels the purines and pyrimidines as it does in other organisms. In the amphibian embryos, however, the quantitative distribution of radioactivity is markedly different from that in the ribose nucleic acidrich bacteria or in slowly growing mammalian tissues. Early in amphibian development the desoxynucleic acid, which is thought to be especially associated with the

genetic material of the cell, has a much higher specific radioactivity than the ribose nucleic acid. As the embryos progress in development, this difference becomes less apparent. In young larvae a nucleotide has been found which has a transient existence but which appears to give rise to certain of the "permanent" nucleotides characteristic of adult tissue.

Storage of Glutamate by Staphylococcus Cells

Certain gram-positive bacteria can store amino acids in their internal environment at concentrations greater than that in the growth medium. Observations of similar storage in higher animals suggest that this process may have general metabolic significance.

E. F. Gale and his co-workers have carried out a series of intensive investigations of the storage process in Staphylococcus (Micrococcus pyogenes var. aureus Duncan) and Streptococcus faecalis, using specific decarboxylase preparations for the estimation of the amino acids. They found that the concentration of glutamate inside the cells, which would not wash out in short exposure to buffer or water, was as high as 150 µmol per milliliter of cells. This corresponds to more than 10 per cent of the dry weight of the cells. The internal concentration of glutamate was in some cases 50 times the external concentration. If other amino acids were introduced in addition to the glutamate, the storage was reduced and in some cases small peptides appeared in the external medium. If a complex mixture of amino acids was added, the storage fell nearly to zero and there was an increase in the combined (protein) glutamate. As a result of this series of studies Gale has suggested that there exists an energy-linked transfer of amino acids across the cell wall.

The investigations of the storage of radioactive glutamate carried out in this laboratory revealed that the cell membrane was freely permeable to glutamate and therefore did not support this hypothesis. The permeability of the cell membrane was most clearly shown by the following type of experiment. Staphylococcus cells were presaturated by incubation with glucose and glutamate to reduce subsequent storage, washed thoroughly in the same concentration of buffered glutamate, and centrifuged. The resulting pellet was then suspended in about its own volume of a radioactive glutamate solution of the given concentration, and immediately centrifuged. It was observed that about onethird of the radioactivity remained in the cells. The cells were then suspended in a large volume of the same concentration of glutamate and again centrifuged. This time most of the radioactive glutamate appeared in the supernatant fluid.

It is possible to calculate the fraction of the volume of the cells which, in rapid communication with the external medium and having the same concentration of glutamate, would explain these results. For purposes of discussion, this fraction of the cell volume may be called the "water space," even though the actual mechanism is probably more complex than is suggested by this term. In these experiments the "water space" proved to be 35–45 per cent of the cell volume, in agreement with the values obtained with the sulfate ion (40 per cent).

It has been observed that there is a rapid exchange between the stored and external glutamate at 30° C, which is practically halted at 0° C.

An attempt has been made to determine, by means of paper chromatography, whether any chemical change occurs during the storage process. When the storage occurs during incubation with glucose and radioactive glutamate, the radioactivity appears (in a 50 per cent alcohol extract) as glutamate only, but when the storage occurs in the absence of glucose the radioactivity appears as glutamate and a substance which behaves on the chromatogram like aspartate. The magnitude of the quantity stored (16 per cent of the dry weight of the organism as glutamic acid in one case) and the permeability of the cell wall show that the storage mechanism is an internal process of the cell.

Manganese

Another approach to the understanding of the relation between nucleic acids and protein synthesis can be found in the study of those abrupt changes in the metabolic activities of cells which are called mutations. Dr. Demerec, of the Department of Genetics, has found that manganous ions are effective in producing mutations particularly when the cells are subjected to certain washing procedures (see reports of the Department of Genetics, Year Books Nos. 49, 50, for 1949–1950, 1950–1951). Studies carried out last year showed that the manganese competes with other ions such as sodium and magnesium for adsorption sites in the cell and that some of the effects of the washing procedures can be interpreted in terms of this competition. Adsorption of manganese, however, is only one of several conditions which are required for the production of the mutation selected for study, namely, a back mutation which is the reversion from streptomycin dependence to nondependence, as observed by Dr. Demerec. Consequently, we have continued to study the binding of manganese and its effect on the metabolic activities of the cell in an effort to correlate the biochemical effects with the mutation.

It was found that manganese was bound by the cells even in concentrations of so-

dium or magnesium which were sufficient to prevent reversions (back mutations). This indicates that only some of the possible adsorption sites are connected with the production of reversions. Further experiments on the binding showed that the manganese was more tightly bound (i.e. less was lost during subsequent washing procedures) when the conditions were favorable for the production of reversions. In one set of experiments the manganese was adsorbed by the cells at 3° C; one-half of the cells were then incubated in distilled water at 3° C, while the other half was incubated at 37° C. Subsequent tests showed that the manganese was more tightly bound when the incubation in water was carried out at 37° C. Previous tests had shown that the 37° C treatment was necessary to produce reversions. Consequently, it seems possible that the same chemical change which produces the reversion also causes a tighter binding of the manganese.

Few reversions were observed when the cells were treated with manganese at 3° C even though they were immediately brought up to 37° C after plating on broth agar plates. In an effort to determine why the broth stopped the reaction which could occur in distilled water at 37° C, the losses of manganese were studied in various fluids. Broth was found to be much more effective in removing manganese than synthetic medium, sodium chloride solution, or water. It appears that the manganese is actively removed from the cells by the metabolic processes which are initiated by placing the cells in broth.

This result is undoubtedly connected with the higher uptake of manganese at 3° C than at 37° C. Metabolic poisons (dinitrophenol or Fe⁺⁺) also increase the uptake of manganese, and there is an inverse correlation between the uptake of

manganese and the uptake of sulfur which was used to measure the rate of protein synthesis.

It is interesting to note that the conditions which were found by Dr. Demerec to be favorable for reversions—i.e., cells grown in broth, resting cells, cells washed in 0.3M NaCl—are all conditions which tend to reduce the synthetic activity of the cells.

It appears, therefore, that the manganese is adsorbed on the critical sites only when magnesium has been removed and the synthetic activities of the cell are reduced to a low level. As a subsequent step a change occurs, possibly a change in the valence of manganese, which results in a tighter binding of the manganese and produces the end result of reversion.

As a second approach we have observed the changes in the metabolism of the cells produced by various treatments. Washing in 0.3M NaCl results in a pronounced magnesium deficiency which reduces protein synthesis (as measured by the incorporation of radioactive sulfate into proteins) by a large factor (10–30). If magnesium is restored, the cells appear unaffected. Distilled-water washes are not so effective in removing magnesium, but do remove free amino acids, glutathione, and other components of the cell.

Treatment with manganese at 37° C, as compared with treatment with magnesium, results in lowered synthetic activities of the cell as measured by the incorporation of C¹⁴O₂ into proteins and nucleic acid.

During the year Dr. B. W. Catlin, Marquette University School of Medicine, made similar measurements on the effect of Fe⁺⁺, which also induces reversions. In all cases the treatment which produced the greatest yield of reversions also produced the greatest impairment of the synthetic activities of the cells. In the case of iron, the temperature effect is different and oxygen plays an important role, lending support to the hypothesis that a change of valence is involved.

OPERATIONS AND STAFF

Co-operative Work of the Department

In accordance with the Institution's established policy of co-operation, we have carried on work with representatives of the National Institutes of Health (including the National Cancer Institute), National Bureau of Standards, Department of Defense, Geological Survey, Massachusetts Institute of Technology, Associated Universities, Catholic and Cornell Universities, University of Virginia, and commercial organizations. This collaboration has been extended abroad by inviting investigators from Denmark, England, India, Norway, Scotland, and South Africa to spend varying amounts of time at our laboratory, often on fellowship appointments. One staff member spent several months in Texas lecturing at the University of Texas, and assisting in the initial phases of a biophysics program at Rice Institute.

Our joint seismic program with the Geophysical Laboratory has been continued, as well as the joint investigation concerned with age measurements on igneous intrusives. We are indebted to the Navy, the University of Washington, and a number of mining companies for assistance in the seismic program in Minnesota and the Puget Sound areas, and to the Geological Survey for help with the mineral age determinations.

We are indebted also to the Board of Education of Montgomery County, Maryland, for permission to place one of the trailers housing ionospheric equipment on the Damascus High School grounds.

Harvard University and the National Bureau of Standards have kindly loaned apparatus and given advice that has been invaluable in the radio astronomy program.

The observatories at Huancayo, Peru; Christchurch, New Zealand; Godhavn, Greenland; Cheltenham, Maryland; and Climax, Colorado have again aided the cosmic-ray program by continuing to operate the cosmic-ray meters.

A letter contract was made with the Atomic Energy Commission for the loan of a small quantity (micrograms) of U235 for use, by isotope dilution, in measuring the ages of igneous intrusives and other minerals, especially in Pre-Cambrian rocks. Other government contracts (without subsidy), as mentioned in last year's report, have been continued for investigations of the earth's crust and cosmic rays. One staff member has been engaged in research work for the government during the entire report year, one since September 1951, one until his retirement date September 30, 1951, and one to the date of his resignation April 1, 1952, all on full-time leave of absence. Others have acted as consultants for brief periods.

We are happy to record our thanks to the Office of Naval Research and the U. S. Coast Guard for carrying out the program of explosions in Puget Sound, including the necessary complicated arrangements with respect to the local fisheries officials. Professor J. E. Henderson and the Department of Physics, University of Washington, gave us a welcome base of operations, and we are particularly grateful to J. A. Anderson, Captain of the USCG Cutter *Mallow*, and his men, and to Lieutenant Commander H. Ridenour of the Thirteenth Naval District, Seattle, for their warm personal interest in our activities.

ADMINISTRATION AND OPERATION

Editing of the *Journal of Geophysical Research* was continued, the Institution providing funds to cover part of the publication costs.

A searchlight building was erected on a farm near Poolesville, Maryland, where experiments will be conducted in conjunction with the searchlight operating at the Derwood Experimental Laboratory.

Lectures

Staff members gave some thirty lectures at scientific gatherings here and abroad in addition to those given at the monthly staff meetings.

Visitors gave the following lectures:

March 6, 1952, "Equation of state at extremely high pressures," H. W. Lewis.

May 15, 1952, "Hydrogen-line radio astronomy in the galaxy," J. H. Oort.

June 19, 1952, "Movements in the upper atmosphere," J. W. Findlay.

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STAFF AND ORGANIZATION

SCIENTIFIC STAFF

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Torreson,‡ E. H. Vestine, G. R. Wait,§
H. W. Wells.

Laboratory and Biophysics: P. H. Abelson, E. T. Bolton, R. J. Britten, D. B. Cowie, N. P. Heydenburg, R. B. Roberts.

Guests, Associates, Fellows, and Visiting Investigators: H. G. Booker, Cornell University; Miss B. W. Catlin, Marquette University School of Medicine; S. Chapman, Queen's College, Oxford, England; W. R. Duryee, National Institutes of Health; J. Dycus, University of Colorado; L. Fagg, Johns Hopkins University; F. J. Feagin, Humble Oil Company; J. W. Findlay, Cavendish Laboratory, Cambridge, England; Miss J. Gillespie, National Institutes of Health; H. F. Johnston, National Bureau of Standards; Soren Lövtrup, Carlsberg Laboratory, Copenhagen, Denmark; P. H. Masson, Humble Oil Company; J. N. Nanda, India; A. T. Price, The Royal Technical College, Glasgow, Scotland; Gunnar Randers, Institutt for Atomenergi, Oslo, Norway; Mrs. I. Z. Roberts; B. F. J. Schonland, University of Witwatersrand, Johannesburg, South Africa; F. L. Talbott, Catholic University; G. M. Temmer, National Bureau of Standards; R. E. Trumble, Jr., Catholic University; T. W. Tuve, University of Colorado.

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Administrative: M. B. Smith, W. F. Steiner.

Office and Clerical: Miss I. Debber, Mrs. E. D. Deininger, W. N. Dove, W. C. Hendrix, Mrs. M. E. Hose, Miss J. S. O'Connor, Miss H. E. Russell, Mrs. F. B. Silberstein, L. Tupler, Mrs. E. A. Walck, Mrs. M. A. Williams.

Instrument Shop: B. J. Haase, L. H. Horton, J. G. Lorz.

Research Assistants, Laboratory Assistants, and Technicians: Miss E. Aldous, S. J. Buynitzky, H. E. Cronin, J. B. Doak, E. T. Ecklund, Miss J. M. George, R. E. Hewitt, P. A. Johnson, C. J. Ksanda, C. A. Little, Jr., M. H. Mac-Kenzie, P. F. Michelsen, P. L. Moats,‡ R. W. Reuschlein, Miss M. Sands,‡ W. E. Scott.

Computers: Miss E. Balsam, Miss I. Lange. Maintenance: C. Balsam, C. R. Domton, I. R. Neely, J. J. O'Donnell, E. Quade, M. A. Quade, S. Swantkowski.

Part-Time and Temporary Employees: Twentynine part-time and temporary employees were engaged during the year, usually for short periods, to assist in the office and laboratory work.

^{*} On leave of absence for government work.

[‡] Resigned.

[§] Retired.



DEPARTMENT OF PLANT BIOLOGY

Stanford, California

C. STACY FRENCH, Director

Recently the biochemical investigations have been devoted largely to the photosynthesis and growth characteristics of the green alga *Chlorella* because of its potential value as food for humans or for animals. The large-scale culture of algae has not yet been demonstrated to be commercially feasible, but if it does become so, the benefits following its introduction may be of considerable magnitude.

Although many fields of plant physiology have had direct application to agricultural practice, research on photosynthesis, one of the natural processes basic to agriculture, has had little practical application. It was therefore satisfying and stimulating to see algal culture as an offshoot of photosynthesis research, our narrow field of specialization, being seriously considered by others as perhaps having potentialities for eventually increasing the world's food supply and bringing about food production by new methods less dependent on the variations of climate and soil. The experiments on the nature of photosynthesis were therefore interrupted early in 1951 to concentrate for a time on the problems directly concerned with the technical development of algal culture. It was desired to make available as rapidly as possible such knowledge of the growth rates of Chlorella, particularly under outdoor conditions, as might be useful in the design and operation of the pilot plant for Chlorella culture which was being undertaken for the Carnegie Institution by Arthur D. Little, Inc., in Cambridge, Massachusetts. The immediate objective of our work on Chlorella was to find out how to grow as much material as possible per day

for each unit area of culture exposed to the Though this objective itself has forced the program into a number of different types of experimentation, some of which were of general interest, the experiments at first were mainly intended to solve those problems which it seemed would be likely to arise in the beginnings of pilot-plant operation. As was mentioned in the last Year Book, the experimental work on these problems soon showed the need for more fundamental understanding of the factors influencing algal growth rates. Thus much of the experimental work of this year came to bear more basically upon the physiology of algae rather than being restricted to measurements of growth rates under conditions similar to those expected to prevail in the actual practice of large-scale culturing. Two fundamental problems, one of photosynthesis and one of the physiology of cellular growth, appear to be in need of solution in order to provide the basic information necessary to devise the means for producing any large increase in the growth rate of outdoor algal cultures. The first of these problems comes from the fact that algae do not utilize light as bright as full sunlight with the high efficiency characteristic of their growth in weak light. Attempts to improve the efficiency of utilization of bright light by exposing the algae to it intermittently have shown the need for a more precise knowledge of the factors controlling the rate of photosynthesis in light of various intermittence patterns and intensities. The other problem is to find out the nature of and the factors influencing the process that limits the growth

of cells when they are given an opportunity to carry on photosynthesis at a rate greater than their capacity for growth.

We have had some occasion, in connection with the Chlorella experiments, to consider the much discussed relations between fundamental and applied science. Clearly there is a two-way interaction between them: just as achievements in science may lead to technical progress, so may the study of a practical problem point out gaps in the underlying science. Thus each of the contrasting aspects of science, the basic and the applied, may serve to stimulate activity in the other. Useful consequences of each small advance in the deeper understanding of the workings of atoms or of living organisms usually cannot be foreseen before the advance is made, and often are not realized until long afterward. Any particular bit of research may remain buried in the great mass of scientific publications, or may equally well eventually open the way to a big step forward. It is, however, evident that the correlated sum total of such studies provides the fundamental basis for the future intellectual and material evolution of civilization. And yet, even though the aesthetic and utilitarian worth of an integrated body of scientific knowledge and theory is obvious, the men who spend their entire lives adding little increments to such a structure often may question in their own minds the ultimate value of their particular contributions when these seem to be only remotely related to ordinary human needs.

The participation in the attempted practical development of a field in which the fundamental scientific facts are still not well known has brought clearly to the mind of each individual the fact that the advances of major significance in practical applications depend for their success more upon adequate theoretical understanding of the process involved than upon the

empirical findings necessarily characteristic of much applied research. The developmental research linking theory with practice is an essential step of such obvious importance in establishing a new process that it is in little danger of being slighted.

The return to the kind of work which has for its end the deeper understanding of natural processes and their correlations with one another is being made with the belief that, whereas specific technical problems may often be adequately met by empirical methods, a more enlightened comprehension of the fundamental aspects of the relevant science is likely to lead, in the course of time, to far more valuable practical developments at the applied level.

With the realization of this fact, the opportunity is again being grasped of concentrating on investigations of the basic nature of photosynthesis and growth.

Clearly the responsibility falling upon investigators engaged in this kind of research is to see that the work planned does in fact aim toward the clarification of questions that are really fundamental to the subject, and not only of interest in the fringes of knowledge surrounding a basic question. Fortunately, the fundamental questions are frequently the simpler ones to formulate, however hard they may be to solve. Because of the difficulties in approaching the basic questions of photosynthesis, a great deal of work has been done on less important subsidiary, though often complex, problems. The resulting accumulation of information has come to be of great volume; some of it is useful, some of it trivial, and some of it confuses the basic issues with elaborate hypotheses having little bearing on reality. To pick out from this mass of knowledge those parts that describe in simple terms the components and mechanism of action of the plant's photosynthetic apparatus, and from this sound base to carry further researches

into the heart of the subject with well defined objectives and by whatever experimental methods need to be devised, is clearly of importance. This can be done with the confidence that the growth of the science will give clearer descriptions and comprehensions of the natural processes; and, in addition, unforeseeable practical benefits are likely to follow in the course of time.

To collect and correlate in usable form the experience of different laboratories working on large-scale culture of algae or on closely related problems of algal physiology and growth, the Institution is preparing a monograph under the editorship of Dr. John S. Burlew, with the title "Algal Culture: From Laboratory to Pilot Plant." This book will include chapters written by workers in the United States, England, the Netherlands, Germany, Israel, Japan, and Venezuela, as well as reports covering the design and operation of the pilot plant by Arthur D. Little, Inc., and the relevant investigations of this Department.

One of the previously mentioned basic difficulties in achieving efficient capture of the energy of sunlight by an algal culture is the inability of algae to use bright light as effectively as they do dim light. It has long been known that bright light given in short flashes results in more photosynthesis per unit amount of light than does light of the same high intensity given continuously. It is hoped to utilize this effect for increasing algal growth rates in sunlight by making the light falling on a particular cell in a dense algal culture intermittent by means of high turbulence in the culture. For this reason the intermittence effect has been investigated in two ways: by re-examining the effect of flashing light on photosynthesis in Chlorella, and by making direct measurements of the growth rates of a dense suspension of Chlorella

in bright light with various degrees of turbulence.

In the flashing-light experiments on photosynthesis in *Chlorella*, appreciable extensions of the previously measured ranges of intensity and of the flashing schedules were used. The efficiency of light utilization for various light and dark periods at different intensities and temperatures was determined. This study later led to some new interpretations of the kinetics of the reactions of photosynthesis.

The turbulence experiment was a series of direct measurements of the growth of a dense suspension of *Chlorella* in bright light, using various degrees of turbulence in a specially constructed culture chamber. This experiment showed a 71 per cent increase of growth with the highest turbulence used, as compared with the lowest at which an adequate amount of carbon dioxide could still be supplied.

A discovery that may have value in the utilization of sunlight by algae was made by Mr. Constantine Sorokin at the University of Texas, working under the direction of Professor Jack E. Myers, a Visiting Investigator of this Department. An algal strain, apparently a form of Chlorella, was found that has an optimum growth rate at 39° C, a much higher temperature than that which is optimum for most algae previously used for mass cultures. Its ability to thrive at high temperature should greatly reduce the difficult problem of adequately cooling a large outdoor installation. Its potential importance may, however, be even greater in other ways than this, because of its ability at the higher temperature to use bright light with greater efficiency than does the common variety of Chlorella. In continuous bright light, very thin suspensions of the new strain at high temperatures grow at twice the rate characteristic of the ordinary Chlorella at its own optimum temperature. There

still remain a number of questions about the growth rates in dense suspensions to be answered before this strain can be recommended for a pilot-plant trial. These experiments have nevertheless shown the advantages to be gained by intentionally selecting from nature the alga best fitted for a prescribed environment, and have made available for further experimentation an alga giving higher rates of photosynthesis for a given amount of living material than has any plant previously measured.

Dr. Robert W. Krauss, of the Botany Department, University of Maryland, who is a Fellow of the Institution, has, under a co-operative arrangement with his University, been engaged in the large-scale culture of a number of strains of the alga Scenedesmus. This genus is of particular interest because of its reputedly high content of sterols of a type possibly useful as starting material for the synthesis of cortisone. Large-scale cultures in special vats and in large bottles have produced approximately a pound dry weight of each of eight strains. In addition to sterol analyses, investigation of the types and quantities of organic phosphates in the different strains is under way.

Earlier experiments on the favorable growth temperatures for Chlorella, performed generally under constant illumination and at low intensity, had shown an optimum temperature of 25° C. A series of measurements of Chlorella growth in sunlight at controlled temperatures showed that 33 per cent higher yields could be obtained by keeping the culture at 30° C during the day and cooling to 20° C at night. These experiments strongly indicate that appreciably higher yields might also have been obtained in the rocking tray and presumably in the pilot plant if those cultures had been at an optimum temperature regime for day and night. Outdoor cultures of Chlorella grown with skylight or shade produced less than cultures in sunlight. The yield was roughly proportional to the light intensity.

Chlorella cells when illuminated grow in size and eventually divide into four or more smaller cells. The environmental factors that control the time of cell division and that of cell enlargement were found to be very different. Increase in cell size occurs during the day and is independent of night temperature. Cell division, however, may be made to occur at night, during the day, or during both day and night by appropriate night temperature control.

It is evident that, ordinarily, neither the rate of cellular division nor the rate of photosynthesis is the limiting factor in *Chlorella* growth. The identification and clarification of the particular process that determines the maximum rate of algal growth when conditions are most favorable remains a basic problem for the future.

Studies of the nitrogen metabolism of *Chlorella* have shown the advantages of using urea as a nitrogen source in large-scale cultures. Growth rates are higher with urea than with potassium nitrate, the acidity of the culture does not change as the nitrogen from urea is used up, and much more nitrogen can be provided at one time than with either nitrate or ammonia without leading to decreased rates of growth.

The settling rate of the commonly used strains of *Chlorella* was measured to provide the required data for engineering estimates of the feasibility of harvesting or concentrating large-scale cultures in settling tanks.

In order to compare the yields of organic matter for a whole year of a common crop grown under very favorable conditions with the yields found for algae, the experiment with two strains of rapidly growing range grass was continued. The average daily yields over a year were about 7 and 8 grams per square meter per day, which is roughly three-quarters of what was obtained from algal cultures. It should, however, be pointed out that the composition of algae is very different from that of grass in that the weight of algae is largely made up of the more useful proteins and fats rather than of cellulose, which accounts for a large part of the weight of the grass.

The influence of carbon dioxide concentration on the growth rate of *Chlorella* was determined by a new method. It was found that very low percentages of carbon dioxide may be utilized without lowering the growth rate, provided that adequate quantities of the gas diluted with air are available and that very good mixing of the gas with the liquid is assured.

Although those aspects of photosynthesis that might bear directly or indirectly upon algal culture received most of the attention of the Biochemical Investigations group, a small amount of progress was also made through other experiments on the photosynthetic apparatus in plants. The fluorescence spectroscopy of chloroplast pigments has been extended to a study of the energy transfer from chlorophyll b to chlorophyll a in concentrated solutions of the pure pigments, since this transfer is believed to be of appreciable significance in ordinary photosynthesis. Some fluorescence spectrum measurements have been made with suspensions of live Chlorella cells, although the minor experimental difficulties caused by selective reabsorption of the fluorescent light within the suspension have not yet been overcome. The purpose of these measurements, if they can be successfully completed, is to bring new data to bear on the old question as to just how much of the chlorophyll a fluorescence, and, by analogy, how much of photosynthesis, in a living cell is due to absorption of light by each of the other pigments present.

The last stage in the series of reactions leading to chlorophyll formation is the transformation of protochlorophyll, a pale green material, to the darker green chlorophyll. The investigation of this purely photochemical reaction, which takes place only in more or less intact plant tissue, has been continued, and efforts are being made to find methods of extracting the protochlorophyll in its natural state of combination with other substances, probably proteins. Protochlorophyll removed from its naturally associated carriers by solvent extraction or by heat denaturation is no longer capable of being converted to chlorophyll by light. This conversion process is probably of a purely photochemical and nonenzymatic nature, yet it does depend very strikingly upon the maintenance of the relation of protochlorophyll to its protein carrier. Studies of this protochlorophyll-protein relation as it bears on the ability of protochlorophyll to be transformed to chlorophyll may reveal a great deal about the state of combination and mechanism of action of chlorophyll in living material, which is a question of major importance in understanding the plant's photosynthetic system.

The curve analyzer has been expanded so as to be more generally applicable to different problems, and its reliability has been improved. Some construction work remains and minor difficulties still need attention, but it is now usable most of the time. It has turned out to have many applications to the work of the laboratory in ways that were not at first anticipated.

Eight years ago the Experimental Taxonomy group started an investigation of the relationships among some of the species of the bluegrass genus *Poa*. The plan was to attempt the crossing of species widely different in their taxonomic classification

and in their climatic adaptations. It was hoped that the principles of plant evolution that have been the concern of the Experimental Taxonomy group during the past two decades might be applicable to grass breeding and that the desirable characteristics of very distantly related species of Poa might be combined into constant hybrids. Such factors as growth rate, size, resistance to drought and pests, seasonal dormancy, and particularly the ability of a favorable strain, once developed, to reproduce itself prolifically without danger of losing its identity by crossing with its parent species were thought to be capable of control by appropriate breeding and selection procedures.

The bluegrass species chosen for these crosses were those that largely produce their seeds by asexual means. Very few attempts at crossing plant species of this type had been undertaken by plant breeders because of the extremely small proportion of hybrids that result from such attempts. When crossing of distantly related species can be done successfully, the way will be open for the introduction of a much wider range of characters than can be obtained within a single species. For practical reasons, most of the previous work on rangegrass breeding had been confined to crosses within, rather than between, species. At the time this investigation was started there was considerable question as to the possibility of obtaining a sufficient number of hybrids by intercrossing the asexual species of Poa. Once hybrids were produced from the very small percentage of crossable flowers, some of the resulting hybrids were themselves expected to be stable by reason of the very property that makes the crosses so difficult to obtain. In nature, competition and evolutionary selection have been operating through past ages to select those species particularly well adapted for survival in each specific local

environment. Nature, however, has not necessarily provided means for selecting grasses which couple this environmental adaptation with a capacity for producing maximum amounts of food for cattle.

A review of the necessary steps involved in producing new grass strains, of the time scale of the program, and of the results obtained up to the present time, as well as an appraisal of the future possibilities of this work, is given here briefly and in detail later in this report.

There is a definite limit to the environmental range for which any particular strain of grass is adapted. Even moderate climatic differences between areas not too far removed from each other may require that entirely different grass strains be used. There is, therefore, no simple single solution to the problem of finding suitable range-grass strains. Many different grasses having various combinations of inheritances are required in order to obtain at least one good type for each main variety of climate. This means that the testing must be done on a scale comprehensive both as to numbers of plants and as to the environments in which they are tested. It is essential also that the new hybrids be simultaneously compared with agricultural strains already established. Furthermore, annual variations in average rainfall and temperatures make it necessary, particularly in the later stages of a test program, to carry the observations through several years.

The procedure followed has been to accumulate strains of potential parent grasses that have many different desirable characteristics, to intercross the best of these in various combinations, and to select the resulting hybrid seedlings among the great numbers of progeny obtained.

The more vigorous hybrids were selected for offspring tests to determine whether they were able to reproduce themselves

asexually like their parent species. Sexually reproducing interspecific hybrids are extremely variable and produce many weak offspring. Asexual production of seed, technically called apomixis, perpetuates the mother plant, in this case the hybrid, unchanged. It is therefore of considerable practical significance. Some hybrids already were constant or apomictic in the first generation, but others did not become so until the second or later generations. The only way to see if a plant is apomictic is to grow its offspring. The testing is of necessity slow, since Poa normally does not flower before its second year, and the full characteristics of the new apomictic strains are not known before the plants have been tested for several years.

A simplified picture of the development of the Poa program is given below in table 1 to show the flow of plants through the various stages of breeding and testing. The first wave of approximately 300 firstgeneration hybrids was obtained from the crossings performed in 1943-1945. The breeding phase of this experiment includes tests on vigor, seasonal periodicity, disease resistance, apomictic production of seed, range of tolerance to climates, and so on, and notes taken on the plants in many contrasting climates. During this stage of the program the plants were grown spaced in nurseries. These preliminary tests in spaced plantings led to the discovery of approximately 30 promising apomictic hybrid strains of Poa. The next stage in the testing program involves sowing the 30 promising strains in replicated plots under normal range, hay, or pasture conditions in dense plantings; this stage will begin in the fall of 1952. The strains which perform best under these conditions will be used for seed increase.

From the results now available it seems clear that certain new combinations of the original parent species and of some of the new constant hybrids may be expected to give even more favorable strains than those now at hand. Therefore, another series of crossings were performed last year and about 400 new first-generation hybrids were obtained from these crossings.

The scope of the *Poa* experiments is to some extent indicated by the fact that more than 150,000 individuals have been studied. The two batches of approximately 700 first-generation hybrids were obtained by selection from approximately 88,000 nonhybrid plants. The 30 constant hybrid lines now suitable for plot testing were picked from about 50,000 plants of hybrid origin which represented several generations and which were thoroughly studied for several years in a series of contrasting environments. These small odds are not unusual in plant breeding utilizing crosses between species.

In order to have some likelihood of producing a grass of practical value for range conditions, it is necessary to carry on many diverse series of tests on large numbers of hybrids. Only by the close co-operation of the U.S. Soil Conservation Service and of several European experimental stations have adequate diversity and magnitude of testing been possible. The Soil Conservation Service made available mature plants of their more promising nonhybrid strains for the initial selections and crossings. It continued the co-operation by growing large cultures of the hybrids in direct comparison with many different grasses in the contrasting environments of its nurseries at Pullman and Bellingham in Washington, and at Pleasanton and San Fernando in California. When a superior strain has been produced and its superiority has been proved, the Service has facilities for initial seed increase, registration, publicity, and distribution to ranchers and seed companies for growth of certified seed.

The Poa experiments have provided an

excellent group of plants for a study of the functioning of apomixis and high polyploidy in evolution. They have clarified the taxonomic, ecologic, and cytogenetic properties of the natural species of *Poa* and their hybrids, and have shed light on the evolutionary history of the genus. The practical importance of the bluegrasses has made possible much more widespread testing by many interested agencies and

individuals than would have been possible for any plant of purely scientific interest. Whatever the practical value of any of the constant hybrid strains of bluegrasses may turn out to be, the program has in both theoretical and practical terms already shown that asexual grasses of different species can be bred to produce constant hybrids in reasonably large numbers, and that such hybrids behave like new species.

PERSONNEL

BIOCHEMICAL INVESTIGATIONS

Staff: C. Stacy French, Director, Harold W. Milner, James H. C. Smith, Herman A. Spoehr, Chairman Emeritus, Violet K. Young.

Visiting Investigator: Jack E. Myers, Professor of Zoology, University of Texas.

Fellows: Bessel Kok, Robert W. Krauss.

Research Assistants: Edwin A. Davis, Jean W. Dedrick.

Technical Assistants: Donald R. Bellis, Richard M. Cook.

EXPERIMENTAL TAXONOMY

Staff: Jens C. Clausen, Paul Grun, William M. Hiesey, David D. Keck.¹

Guest Investigators: Friedrich Ehrendorfer, Axel Nygren.

Stanford University graduate students associated with Department: Henry J. Thompson, Robert K. Vickery, Jr., George H. Ward.

Research Assistant: Malcolm A. Nobs. Technical Assistants: Robert W. Ayres, Oliver W. Norvell, Edward L. Triplett. Gardener: Wesley B. Justice.

RESEARCH ASSOCIATE

Ralph W. Chaney, Professor of Paleontology, University of California, Berkeley.

DEPARTMENT SECRETARY Wilbur A. Pestell.

Mechanical Engineer Louis R. Kruger.

¹ Resigned December 31, 1951.

Custodian

Richard P. Ludolph, Jr.

During the year Dr. Herman A. Spoehr, Chairman Emeritus, served as president of the Pacific Division of the American Association for the Advancement of Science, giving the presidential address, "Society in the Grip of Science," at the meeting at Corvallis, Oregon, on June 18, 1952.

Dr. William M. Hiesey, on invitation, presented a paper at the Colloque International sur l'Evolution chez les Plantes in Paris, visited a number of European laboratories, and took notes on the growth of the Department's range-grass hybrids being tested at different experiment stations in Denmark, Sweden, Norway, and Scotland. Dr. Hiesey will also attend the symposium on evolution of the Society for Experimental Biology at Oxford, England, July 7 to 11, 1952.

Dr. Friedrich Ehrendorfer, of the University of Vienna, Austria, came to the Department on May 9, 1952, to spend four months as a guest investigator in the study of *Achillea*.

Dr. David D. Keck resigned as of December 31, 1951, after twenty-three years with the Institution, to continue his work as head curator of the New York Botanical Garden, having gone to the Garden last year on leave of absence from the Institution.

EXPERIMENTAL TAXONOMY

During the year 1951-1952 progress has been achieved along several fronts of the experimental studies on the evolutionary relationships of plants. A major new venture has been started in the launching of investigations on the comparative physiology of contrasting climatic races of the same or closely related species. These studies are aimed at determining the fundamental physiological basis of natural selection in higher plants. Biosystematic researches were successfully concluded by Robert K. Vickery on the genus Mimulus, Henry J. Thompson on Dodecatheon, and George H. Ward on the Artemisia tridentata complex. These three studies were subjects of doctoral theses submitted to Stanford University.

SURVEY OF THE RANGE-GRASS PROGRAM

JENS CLAUSEN, WILLIAM M. HIESEY, PAUL GRUN,
AND MALCOLM A. NOBS

The range-grass program was started in 1943 on the basis of the idea that it would be possible to intercross apomictic species of bluegrass and thus combine the inheritances of contrasting species from contrasting climates into stable hybrids with new qualities. In this manner it was hoped to draw from the pools of genes within a large sector of the genus rather than to be circumscribed by the genetic resources of the individual species. By utilizing asexual seed production it was also hoped to avoid some of the dangers of weak offspring after interspecific hybridization and to shorten the time normally required for the development of new self-reproducing strains.

At the start of the investigation relatively little was known about the characteristics, ecologic tolerances, seasonal periodicities, and life cycles of the wild species of *Poa*. The species of agricultural significance were known to be perennial, but it was not

known how soon they would flower after being sown under California conditions. It is now known that most species require two years for this development, which is the minimum time required for preliminary observation; for tests on performance it is necessary to study a given strain over a longer period. The time factor is therefore an important limitation in the development of a new strain. The fact that Poa is apomictic helps to speed the development of new strains, because vigorous truebreeding individuals from which new strains can be established have been obtained in the first and second generations following interspecific hybridization.

The tests for apomixis of hybrid individuals have been performed by testing the constancy of their progeny derived by open pollination. Because large numbers of individuals were involved, it was found impracticable to apply controlled pollination. Also, apomictic hybrids remain true to type irrespective of the pollen available to them.

In the case of sexual hybrids it was probably fortunate that the procedure of open pollination was followed, because thereby the *Poa* hybrids were exposed to all the pollen of different species and hybrids available on the field at the time of flowering, and this permitted a strong natural selection among fertilizing pollen. The results of the open pollinations indicate that the hybrids which were not apomictic were either fertilized by their own kind of pollen or backcrossed to their parent species.

Some interspecific *Poa* hybrids are already apomictic in the first hybrid generation, but their apomixis cannot be discovered before tests on the constancy of their progeny have been performed as described above. In *Poa* more than half the F₁ hybrids between two apomictic species are sexual and segregate in the F₂. Sometimes such a segregation proves to be ad-

vantageous by providing the opportunity for selection among a segregating progeny in which many recombinations of the parental characters occur. By selecting the agronomically most desirable F₂ individuals for apomictic types one may be reasonably certain that any constant F₃ type will be a desirable strain.

After a new line has been established there remains the problem of finding the range of environments for which it is adapted. The agricultural value of *Poa* facilitates such tests, because agencies such as the Soil Conservation Service and many experiment stations seeking new range and forage grasses are willing to test the strains in the environments available to them.

The progress made to date would have been impossible without the sustained cooperation of the Pacific Coast Division of the U.S. Soil Conservation Service. Under the supervision of Dr. A. L. Hafenrichter, chief of the Nursery Division of the Pacific coast region, the Service supplied mature plants for immediate crossing, and seeds of strains which its staff had collected over a period of years and on which preliminary tests had already been made. A considerable part of the second hybrid generations were grown at the Soil Conservation nurseries at Pullman, Washington, and at San Fernando, California. In the later stage the Service devoted much nursery space and effort to the testing of progenies and strains at their major nurseries. Dr. Hafenrichter and Mr. John L. Schwendiman, manager of the Pullman nursery, aided our staff in the selection of strains. Other members of the staff of the Soil Conservation Service at the various nurseries have extended every help and courtesy throughout the years. We are especially indebted to Mr. Donald Douglas and Mr. Richard Adlard at Pullman, Mr. W. E. Chapin and Mr. Lynn Gunther at Bellingham, Mr. Harold W. Miller, Mr. Oswald Hoglund,

and Mr. W. T. McLaughlin at Pleasanton, and Dr. Paul Lemmon, Dr. Lowell A. Mullen, Mr. Jack Woods, and Mr. Dirk Vanderwal at San Fernando.

Table 1 summarizes the development in time of the various stages in the Poa program. Seven crossings were made in 1943, but the major part of the hybridizations could not take place before 1944 and 1945, after the various species had been assembled at Stanford. The first F1 hybrids were planted from 1944 to 1946 at Stanford. Second-generation progeny of the combination Poa scabrella × pratensis, which blooms the first year, were planted at Stanford and Pullman in 1946 and at Stanford, Pullman, and San Fernando in 1947 through 1949. Progeny of the third generation were first planted at Stanford and Pullman in 1948 and 1949, and of the fourth generation in 1951.

The testing of parent species at the transplant stations began in 1944, and clones of the first hybrids were planted at Stanford, Mather, and Timberline in 1945. The first apomictic F₁ hybrid strains were discovered in 1946 and 1947. Only the strain from 1947 is still considered to be of potential agronomic importance. It was not until 1949 that the first strains that had been derived from apomictic F2 plants could be recognized. At this time enough apomictic strains were available for largerscale testing of their climatic tolerance in the intercontinental transplant experiment carried on in co-operation with experiment stations in the Netherlands, Denmark, Sweden, Norway, Scotland, and Wales, in addition to the tests at the Soil Conservation Service nurseries in the Pacific coast states and our own altitudinal stations. The selection of strains available at that time was very limited, and much better strains could now be supplied. The intercontinental experiments served a useful purpose, however, because they made possible

 $\begin{tabular}{ll} TABLE & 1 \\ SURVEY & OF THE RANGE-GRASS & PROGRAM \\ \end{tabular}$

	1943	1944	1945	1946	1947	1948	1949	1950	1951	1952
Nonhybrid strains grown		92	160	5			23	14	29	15
No. crossings made	7	24	22						34	
No. seedlings grown		5,200	43,000	15,300						25,000
No. F ₁ hybrid individuals discovered	١	13	184	105						400
No. cloned individuals set at three	:									
transplant stations		32	63	172	155	72		68		
No. hybrid offspring planted at CIW and SCS:	7									
F ₂ , individuals				7,300	2,400	6,100	1,200		450	1,150
F ₃ , individuals						7,900	2,700	2,000	960	2,630
F ₄ , individuals									1,300	
No. individuals tested for tolerance to climate at:										
San Fernando					800	700				1,500
Pullman				300	1,000	2,500	1,700	2,000	2,500	3,500
Bellingham							1,600			600
Edinburgh					2,000		1,200			
Wales							100			• •
Netherlands				• • •			450		• •	
Denmark			• •		• •		2,700	• •	• •	• • •
Uppsala Norway			• •	• •			1,200 3,300			
Pleasanton								1,900		
No. strains tested for tolerance to climate at:		••	••	••	••	••	100	1,500		
Alaska							• •	• •	7	
Southern California				• •	• •	• •	• •	• •	6	10
Cornell University		• •			• •	• •	• •	• •	15	11
South Africa, 6 stations		• •	• •	• •		• • •			• •	11 19
Sweden, 6 stations		• •	• •					• •	• •	19
discovered:										
F_1 type				3	1		5	1	1	
F ₂ type							11	1	11	6
No. apomictic strains selected for plot tests:										
F ₁ type										7
F ₂ type										23

an ecological characterization of the parental species and some of the hybrid combinations. This was reinforced by Hiesey's physiological studies on parental and hybrid strains in 1950 at the Earhart Plant Research Laboratory of the California Institute of Technology, and by the studies of Mrs. G. Juhrén and Dr. F. W. Went on germination and early seedling growth, also at the Earhart Laboratory.

Some of the most promising apomictic types were not obtained until 1951 and 1952 at Pullman. During these two years 18 new strains were discovered, so that some of those previously considered to be of promise have already been superseded by better lines. The Poa ampla-pratensis hybrid of the Palouse Prairie form of P. ampla from Albion, Washington with a form of P. pratensis from a mid-altitude meadow in the Sierra Nevada at Mather has been a favorite combination which is represented by a series of lines that were already apomictic in the F₁. These have two sets of chromosomes from ampla and about one from pratensis. They have short rhizomes and are quite successful at our three transplant stations, but become excessively stemmy except when grown in the central California Coast Ranges, where they are acceptably leafy. The Albion × Mather hybrid population also contains, however, another F₁ type which is composed of one set of chromosomes from each parent. This type has longer rhizomes and is more leafy, but it is also sexual. By selection among the best F2 plants of this type, about 6 apomictic strains of considerable promise were obtained at Pullman. They are more leafy and vigorous than pratensis, and have larger inflorescence and almost as long rhizomes. They bloom several weeks later than pratensis and have bluish foliage indicating inheritance from ampla. Seeds are currently being harvested from these strains, and if

any of the strains produce enough seed they will be included next year in the plot tests, although their tolerance for a range of environments has not as yet been tested.

Up to the present time more than 150,000 Poa plants have been examined and about 700 F₁ plants produced. Approximately 50,000 hybrid individuals of several generations have been studied individually and measured carefully for segregation of characteristics in up to thirteen different environments. At Pullman alone, approximately 13,500 individuals of the hybrid cultures are still being studied. On the background of these studies, between 25 and 30 lines have been selected for further tests in small replicated plots sown according to normal agronomic practices. The exact number of the strains to be tested will depend on their seed fertility in 1952.

Emphasis has hitherto been placed on the genetic analysis and selection of apomictic strains, requiring spaced planting with seedlings planted in gardens from seedling flats. The next step is to test strains in dense plantings with seeds drilled directly into the ground. These plantings will test the ability of the seeds to germinate in the field and to produce in dense plantings. Sufficient seeds for such tests were secured in the summer of 1951 from plantings at Stanford, Pleasanton, Pullman, and Bellingham. A total of more than 30 pounds of seeds were collected from the hybrid lines in 1951, and more will be available from the 1952 crop.

When the data on the performance in the replicated tests have been analyzed, the next step will be to select the few strains that may have passed this test for seed increase on a larger scale and for still more exacting tests in competition with weedy annuals on the unimproved range. It is possible that in final practice it will be found advantageous to grow several strains

together on the range which will be active and bloom at different periods but collectively will form a successful community in competition with less desirable plants. Seed stocks of such strains could be produced separately where commercial seed production is best, and then combined into mixtures tailored for local conditions. It will therefore be of importance to have a number of suitable strains available to meet such demands and to fit the major regions within the western range.

Before a new strain can be introduced into practical use, many steps are necessary. The strains have to be tested carefully for resistance to pests, and their hardiness under grazing and various other range and agricultural practices must be determined. The Soil Conservation Service has facilities for registering and publicizing new strains and for contracting with farmers and commercial growers for the raising and distributing of certified seed stocks.

The seeds of the hybrid strains collected in 1950 were used in establishing new plantings in various parts of the world. Seven strains were sent to Fairbanks, Alaska, under auspices of the Soil Conservation Service. Fifteen strains were planted at Cornell University, Ithaca, New York, at the request of Dr. Sanford Atwood, head of the Department of Plant Breeding. At the request of Dr. L. C. C. Liebenberg, of the Department of Agriculture, Union of South Africa, seeds of 11 strains were sent to Pretoria to be planted at six climatically contrasting stations. Nineteen strains were supplied to Drs. Erik Åkerberg and Axel Nygren, of the Royal Agricultural College, Uppsala, Sweden, to be planted at six stations from southern to northernmost Sweden above the Arctic Circle. Dr. Nygren, who was a Guest Investigator at our Department in 1951, plans to use our strains in crossing

native Swedish species of Poa. Seeds of 10 strains were supplied to Mr. G. Juhrén, of the California Forest and Range Experiment Station, for sowing broadcast on burns in the interior regions of Los Angeles County near Lancaster. Step by step the new hybrid lines are therefore being tested under very different practices and in very different environments of the world, from 45° south latitude to 68° north. The development of satisfactory new agronomic strains is a long and complex process in any kind of plant. It taxes the limits of fundamental knowledge of the biology of plants, and is intimately related to the exacting demands and limitations of management practices.

New Poa Hybrids

JENS CLAUSEN, PAUL GRUN, WILLIAM M. HIESEY, AND MALCOLM A. NOBS

When our first Poa hybrids were produced, in 1943-1944, it was impossible to predict how the genomes of the various species would react when combined into a hybrid. Not even the tolerances and characteristics of their wild parent species were well known. Although a sufficient number of hybrids were obtained in some of the combinations, there were too few in others to assure success against the odds prevailing in interspecific crossing of apomictic species. After the first crossings some additional species and races of potential value were obtained, and others came to our attention as their qualities became known.

In order to explore the qualities of these races and species, especially for the drier and more southern regions, a series of 34 new crossings were attempted in the spring and early summer of 1951. The technique applied was the same as that described in Year Book No. 43 (1943–1944, pp. 72–73), namely, enclosing well developed indi-

viduals of two contrasting species within a pollen-tight cage and mass-pollinating them each morning without previous emasculation.

Approximately 25,000 seedlings germinated in the spring of 1952. At the age of approximately 3 months, seedlings deviating from the maternal type and therefore considered to be possible hybrids were marked for further observation. Later they and samples of the nonhybrids were potted individually. By July approximately 400 hybrid individuals had been recognized from 22 of the 34 crossings attempted, 12 combinations apparently not having produced hybrids. The frequencies of hybrids in both successful and unsuccessful combinations are listed in table 2. This list supplements those in Year Books No. 44 (1944–1945, p. 75) and No. 46 (1946–1947, p. 98), reporting the results of the first group of 53 crossings that led to the selection of the hybrid and apomictic strains discussed above.

Among the new hybrids is a group of 11 individuals having Poa caespitosa Forst. as one of their parents. This species is a vigorous bunchgrass native to Australia and New Zealand. Its relationship to the Poa species previously included in our experiments seems to be rather remote. Like the native California bluegrass, Poa scabrella (Thurb.) Benth., caespitosa is winteractive, but it also remains summer-active. Poa caespitosa requires climates with mild winters and dies after the first winter when planted at our Mather and Timberline stations. Its variability following artificial self-pollination indicates that it reproduces by sexual means.

Another group of 119 new hybrids has the Texas bluegrass, *Poa arachnifera* Torr., as one parent. This grass is a native of the southern Great Plains and is a vigorous and leafy rhizome species that also is both winter- and summer-active. Like *caespi*-

tosa it reproduces by sexual means, but it is dioecious, that is, it has ovaries and pollen on different plants, in contrast with most of the *Poa* species of the northern hemisphere, which are hermaphroditic. *Poa arachnifera* is of much practical significance for the winter range in its area, but its seeds are covered with a thick, woolly mat of hairs, an objectionable character that makes cleaning and sowing difficult.

Previously (Year Book No. 45, 1945-1946, p. 98) crossings were attempted between 56-chromosome arachnifera and two Outer Coast Range races of the 84-chromosome Poa scabrella, but no hybrids were obtained among 2025 seedlings, using scabrella as the female parent. With a peculiar 63-chromosome inland form of scabrella from Lake County, California as the female parent, a single hybrid was obtained among 1151 maternals, but this hybrid was very weak and completely sterile. This year, using a female and partially sexual 63-chromosome plant of the coastal form of scabrella as the maternal parent and a male of arachnifera, 35 vigorous scabrella × arachnifera hybrids were obtained, together with 189 nonhybrid maternal plants of scabrella. Among the hybrids that flowered this first year were both hermaphrodites and females.

A cross between *Poa nervosa* and *P. arachnifera* was successfully made. The female *nervosa* parent was of a strain from southeast Washington composed of both females and males. This cross yielded hybrids almost exclusively, but almost all the 55 hybrids were females and only 2 were hermaphrodites. The hybrids of *arachnifera* with *scabrella* and *nervosa* have much less pubescence on the seeds than does *arachnifera*, but longer and denser hairs than do their slightly pubescent female parents.

The ideal hybrid by which to reduce or

TABLE 2
New Poa hybrids, 1952

=			
	Hybrid combination	No. hybrids	Frequency (%)
1.	Poa caespitosa hybrids:		
	ampla, Albion × caespitosa	4	3.3
	Reciprocals	0	0.0
	pratensis × caespitosa	2	1.6
	Reciprocals	0 3	$0.0 \\ 1.0$
	compressa × caespitosa	0	0.0
	Reciprocal	2	1.8
2	Poa arachnifera hybrids:	2	1.0
۷.	scabrella, $2n = 63 \times arachnifera$	35	16.6
	arachnifera × pratensis	26	83.9
	nervosa, Wallowa Mts. × arachnifera	55	96.5
	arachnifera × ampla-pratensis	2	}
	Reciprocal	1	2.7
3.	Poa scabrella, Paso Robles, hybrids:		
	scabrella \times pratensis, Groveland, $2n = 56$	4	0.4
	Reciprocal	0	0.0
	scabrella \times pratensis, Mono Lake, $2n = 50$	63	7.8
	Reciprocal	0	0.0
	scabrella × pratensis, Las Vegas	25	4.0
	Reciprocal	0	0.0
	scabrella × arida-scabrella	28	1.8
4	Reciprocal	3	3
4.	Poa ampla, Albion × compressa, Crescent Mills:	2	0.2
	ampla × compressa	2	0.2
5	Reciprocals	U	0.0
٥.	arida × ampla, Albion	22	1.9
	arida × ampla, Wenatchee	4	0.7
6	Quadruple hybrids:	-	0.7
٥.	arida-ampla × ampla-alpigena	16	3.0
	Reciprocal.	4	0.5
	Reciprocal arida-ampla × ampla-pratensis	26	3.0
	Reciprocal	4	2.7
	arida-ampla × ampla-pratensis, Condon	3	1.7
	Reciprocal	8?	
	scabrella-pratensis × ampla-alpigena	59	13.1
	Reciprocal	0	0.0
	ampla-alpigena X ampla-pratensis	8	1.7
	Reciprocal	0	0.0
			No.
			maternal-type
7	Unsuccessful hybridizations:		seedlings
٠.	caespitosa × scabrella		43
	Reciprocal		1,128
	Howellii × scabrella, Gold Beach.		958
	Reciprocal		90
	Howellii × pratensis		1,771
	Howellii × Douglasii		660
	Reciprocal		150
	Douglasii \times arachnifera		30
	$arachnifera \times ampla$, Albion		30
	Reciprocal		1,440
	$arachnifera \times scabrella, 2n = 84$		12
	Kelloggii × ampla, Albion		150
	Reciprocal		240
	ampla-alpigena × ampla-pratensis, Condon		44
	Reciprocal		630

remove the seed pubescence of arachnifera would be a cross with the big bluegrass of the Palouse Prairie, Poa ampla Merr., which has no pubescence. This crossing was attempted but failed. Three hybrids were obtained reciprocally, however, in crossing arachnifera with the apomictic P. ampla-pratensis, a constant F1 hybrid containing two sets of chromosomes of ampla and approximately one set of pratensis chromosomes. Poa pratensis crosses readily with arachnifera. This combination was obtained in 1908 by G. P. Oliver and later reobtained by E. Marion Brown. In our present crossing program 26 individuals of arachnifera × pratensis were obtained. None of these, nor any of arachnifera × ampla-pratensis, have flowered the first year.

The hybrids of group 3 of table 2 are thought to be promising in producing grasses for southern dry ranges. They come from crosses between a vigorous strain of Poa scabrella from the dry southern Inner Coast Range of California and three strains of P. pratensis from warm and dry hill-slope habitats. The strain of scabrella used was collected a few years ago thirty miles east of Paso Robles, in San Luis Obispo County, by Dr. David D. Keck of our Department and Mr. H. W. Miller of the Soil Conservation Service. One of the pratensis parents was collected in a dry swale surrounded by yellow pine forest above Groveland in the Sierra Nevada at 3000 feet altitude, another on a sagebrush slope at 6600 feet near Mono Lake, east of the Sierra Nevada on the Great Basin plateau, and the third from near Las Vegas, New Mexico, at 6600 feet at the lower edge of the Pinus scopulorum forest. The last-mentioned strain was collected by Mr. Oliver W. Norvell. The 92 hybrid individuals obtained from these three crossings differ from previous F1's of scabrella × pratensis in having rhizomes,

in being generally more leafy, and in not flowering the first year. The same Paso Robles race of P. scabrella was also crossed with the F_1 of P. arida \times scabrella. The arida-scabrella hybrid is one of the best Poas in the San Fernando Nursery of the Soil Conservation Service in southern California, and remains green there through June. It originated from a cross that contributed two sets of chromosomes from P. arida and one set from a race of P. scabrella from the Outer Coast Range of southern California. This hybrid behaves as a sexual amphiploid species with limited segregation. In the present cross it is hoped to add a set of chromosomes from the Inner Coast Range form of scabrella to an unreduced ovule of the arida-scabrella hybrid. Such a hybrid would be composed of two sets of arida chromosomes and two of scabrella, but the scabrella sets would have come from distinct races of the species. This type of hybrid is difficult to recognize in early seedling stages, but 28 F₁'s harvested on the summer-dormant scabrella were easily identified by their summer activity.

A fourth group of hybrids was produced in an attempt to synthesize a new form combining the big bluegrass, Poa ampla, from the Palouse Prairie with P. compressa. Both are grasses from relatively dry environments, but differ widely in vegetative characteristics. The hybrids bloom late in the season and are of potential value in providing late grazing at a time when few other grasses are available on the western ranges. In the earlier crossings, 9 F1 hybrids of two distinct combinations of ampla and compressa were obtained. These F₁'s were vigorous, but 4 of them were sterile and the other 5 were sexual and produced weak F2 offspring. The progeny of one of these hybrids was followed at Pullman to the fourth generation, but con tinued to segregate. In the present cross it was attempted to combine the Albion form of *ampla* with a form of *compressa* having long rhizomes, originally from Crescent Mills, Plumas County, California; but only 2 hybrids were found among 1725 seedlings of the reciprocal crossings. It seems to be very difficult to produce a successful new combination with these two species.

The crossings listed under group 5 in table 2 were attempted in the hope of combining Poa arida with P. ampla. In 1944 one hybrid each of P. arida \times ampla and its reciprocal were obtained. Both F1's were very vigorous, highly leafy, and highly fertile. Potentially, this hybrid has the appearance of being one of the best combinations for the Great Basin range. The better of the two reciprocals is arida × ampla, which has the longer rhizomes. It has two sets of chromosomes of arida and one set of ampla, whereas the ampla \times arida hybrid has only one set from each parent. Unfortunately, the ampla \times arida hybrid is sexual, and its strongly segregating second-generation progeny were so weak that few plants survived. The arida × ampla F₁ was approximately 12 per cent apomictic and produced F2 progeny with some vigorous plants—the apomicts—plus large numbers of highly variable and generally weak plants, many of which died. Progeny tests on 18 of the better F₂ plants were made at Pullman, but these thirdgeneration plants were found to be either completely sexual, with variable and weak progeny, or only 12 per cent apomictic, like the original F₁. These results indicate that the chances for increasing the degree of apomixis through further selection in this progeny are remote.

Additional crossings made in 1951 for the purpose of producing the desirable *Poa arida* × *ampla* F₁ combination yielded 26 new F₁'s during the current year. They are variable among themselves, as is commonly the case among F₁'s

of apomictic parents. The number of F₁ individuals is large enough to provide a reasonable possibility that at least one of them may produce a bluegrass suitable for the arid ranges. If this method should fail, some of the 50 quadruple F₁ hybrids of P. arida-ampla × ampla-alpigena and P. arida-ampla × ampla-pratensis, Albion-Mather, in group 6 of table 2 should yield desirable lines because the ampla-alpigena and Albion-Mather combinations have so far produced the greatest numbers of apomictic strains.

The 128 quadruple hybrids listed under group 6 of table 2 were produced to determine whether apomictic hybrids behave genetically like natural apomictic species. After two apomictic hybrids are crossed, it is a question whether the recently reestablished apomictic mechanism will again adjust itself to the still newer balance or will disintegrate into the component gene blocks from which the four parental species originally evolved. The best example for such an evaluation is Poa scabrella-pratensis × ampla-alpigena. The parent hybrids of this cross, lines 4711-3 and 4683-1, respectively, have very much the aspects of natural species. They differ from other Poa species, are constant, are fertile, and appear to be adapted for certain specific ecologic ranges.

Line 4711-3, the maternal scabrella-pratensis parent, combines characteristics of the southern California Outer Coast Range Poa scabrella with those of a midaltitude Sierra Nevada meadow form of P. pratensis. Likewise, line 4683-1 combines characters of the Palouse Prairie form of Poa ampla and the Swedish Lapland form of P. pratensis alpigena. Accordingly, the quadruple hybrids of this parentage combine characteristics of four species from very contrasting climates.

Although ampla-alpigena as the female parent yielded hybrids in other combina-

tions, the quadruple hybrids of Poa scabrella-pratensis × ampla-alpigena were obtained only on scabrella-pratensis as the maternal parent. The pollination on the scabrella-pratensis line yielded 59 quadruple hybrids, the highest frequency ever experienced in crossing two apomictic Poas. The nonhybrids of this progeny are uniformly of the maternal type, but the hybrids are very diverse and many of them show characteristics of all four parent species. Two-thirds of the hybrids flowered the first year like the scabrella-pratensis parent, but the morphological influence of ampla-alpigena was very strong. During the current year many of the F₁'s exceed both parents in vigor and in the development of foliage. Taxonomically significant characters vary from plant to plant, so that it appears that the gene blocks are to some extent being broken up and recombined. The quadruple hybrids all have rhizomes and are summeractive, so these characters have not been lost although two of the grandparents were bunchgrasses and one was summerdormant. Otherwise the hybrid population of 59 plants gives the impression that the variation of a good portion of the genus has been thrust into it.

Unsuccessful hybridizations are listed under group 7 in table 2. Outstanding is the failure of *Poa Howellii* to cross with *P. scabrella*, *P. Douglasii*, or *P. pratensis*. *Poa Howellii* is an annual and apparently sexual species occurring in the Coast Ranges from California to Washington. It is sometimes considered to be a relative of the cosmopolitan annual *P. annua* L., which happens to have the same number of chromosomes (2n=28). In gross morphology *Howellii* shows some similarity to forms of *P. scabrella*. The failure to obtain hybrids of *Howellii* suggests that this species belongs to a genetically

very different group of *Poa* from any one of the species with which it was crossed.

Poa Kelloggii Vasey was included in the crossings because it appeared to be a counterpart of *P. pratensis* fitted for the redwood region, although it is morphologically very distinct. It is a tall, leafy, rhizome-bearing 56-chromosome species occurring in ravines along stream banks opening to the coast in the redwood regions, and is therefore not very common. It probably belongs to the Pratenses section of the genus, but unlike *pratensis* it does not appear to cross with *P. ampla*.

Poa Douglasii Nees is another species with which first attempts at hybridization have failed. This failure should not be considered conclusive, because plants of that species were transplanted directly from the dunes along the California coast and used immediately in the attempted crossings. Poa Douglasii is probably conspecific with the more northern P. macrantha Vasey. These two species have been classified in the Pratenses section, which undoubtedly is an error. In experimental cultures they are bunchgrasses and have no rhizomes, but their leafy stems become long and whiplike and bend over by their own weight, rooting at the tips by layering. In dunes these rooting stems become covered by sand and appear to be genuine rhizomes. These two species have male and female plants like P. arachnifera, and are of some importance as sand binders.

It is evident from the study of the crossings performed in 1951 that there are definite limits to the crossability of species within the genus *Poa*. Evolutionarily it is of significance to find where such limits are. Exploration of these limits is complicated by the fact that there are considerable differences between races and even between individuals of species in the ease

with which they are able to cross. At the present time the addition of the inheritances of distinct species into new successful combinations remains as something of an art rather than a predictable science, although to a certain extent the ecologic characteristics of the hybrids can be predicted provided the characteristics of the parents are known.

Apomixis and Variation in Poa Nervosa Paul Grun

In a normal, completely sexual plant a double fertilization occurs: the sexual egg is fertilized and develops into an embryo, and the polar nuclei are fertilized and develop into the endosperm, the food reserves of the seed. In bluegrasses the mechanism of apomixis has resulted in nonfertilization of the egg and development of an asexual embryo which, gene for gene, is a copy of the female parent. The genetic and embryological researches of several Swedish workers, particularly on apomictic *Poa pratensis*, have shown that the fertilization of the polar nuclei is still essential to the development of the endosperm.

Poa nervosa, a common western mountain grass (see Year Book No. 50, 1950-1951, pp. 112-113), has no functional anthers and produces no pollen over nearly all of its range. Embryological study this year has shown that most of its embryosac mother cells are of the apomictic type, inasmuch as they develop following mitotic rather than meiotic cell divisions. Development of an embryo from such an embryosac mother cell would not require that it be fertilized, although the possibility still exists that endosperm fertilization is required. Several other species of bluegrass occur together with P. nervosa, and one of them could supply pollen for such endosperm fertilization. To test this possibility, eight nervosa plants at Timberline were caged in pollen-proof cloth cages previous to flowering. Since the female *nervosa* plants themselves produce no pollen, endosperm fertilization was not possible. The seed set was nevertheless completely normal and the germination of the seeds was high. *Poa nervosa*, then, is capable of normal seed formation without any fertilization of either egg or polar nuclei.

Examination of the chromosomes in the endosperm of Poa nervosa showed that mitosis is highly irregular here. Accordingly, there are variations in chromosome number even within a single endosperm. In five of the six endosperm cells on which accurate counts could be made, the chromosome number was about 120. Since the somatic chromosome number of these Poas is approximately 60, the 120 chromosomes are probably derived from the fusion of two polar nuclei. The sixth endosperm cell was counted in a caged plant and contained about 357 chromosomes, a fact which suggests that it had resulted from a fusion of several endosperm cells.

What types of morphological variation occur in nervosa, considering that only one sex exists? As described in Year Book No. 50, cited above, significant minor variations occur even within a limited area in the Sierra Nevada of California. These variations are of a type that can be demonstrated only by statistical analysis when the plants are grown in replicated plantings. When *Poa nervosa* strains from geographic areas beyond the Sierra Nevada are taken into consideration, even greater differences between forms are found. In the Californian Sierra Nevada the plants grow usually above 7000 feet altitude under lodgepole pines, Pinus Murrayana, in a large continuous area favorable for the species. By contrast, the region of central to eastern Oregon and Washington is traversed by a series of separate mountain ranges furnishing disjunct habitats. Poa nervosa grows here at higher elevations in open pine forests (*Pinus ponderosa*), but only where the competition is not severe. In the region so far examined it appears that the separate ranges support distinct forms. In the Ochoco Pass area of central Oregon, for example, the majority of plants have large exserted, though abortive, anthers. Farther east in Oregon at Dixie Pass, the plants have much smaller, non-exserted anthers. In both areas ligules grow to 2 mm long, in contrast with those of the California plants, which are 1 mm long.

Chromosome counts for Poa nervosa have been made on 20 plants from the Sierra Nevada of central California. These included several forms collected along a transect from White Wolf, Tuolumne County, to Slate Creek Valley, Mono County, a distance of 20 miles, and three forms from Woods Lake, Alpine County, 60 miles north of this transect. Eighteen of the 20 plants had about 63 chromosomes. One of the remaining two, which was morphologically indistinguishable from the others, had 76 chromosomes, and the other, a distinct aberrant from near the Timberline station, had 80 chromosomes. In contrast with this predominance of forms with 2n = 63 found in the central Sierra Nevada, 5 plants from three widely different localities of central and western Washington east of the Cascades had between 81 and 90 chromosomes.

Another form of *Poa nervosa* grows at low altitude in extremely moist, shaded areas west of the Cascade Mountains in the moist forests near the mouth of the Columbia River. In its ecology, its very lush growth of basal leaves, and its possession of functioning anthers it is distinct from the mountain forms. The chromosome number of plants of this type collected near the junction of the Kalama and Columbia Rivers in Cowlitz County,

Washington, was 2n=28. In contrast with the mountain forms, which are predominantly apomictic, the 28-chromosome low-land form produces extremely variable and strikingly sexual progeny. Owing to the pronounced chromosomal difference between these two forms, and particularly to their geographic isolation, it is not believed that the lowland form at present plays any part in the production of variability in the apomictic mountain forms.

Where and how, then, is variation produced among the exclusively female mountain forms? There are at present three known populations which may furnish part of the answer. One of these occurs near Field Springs State Park in Asotin County, eastern Washington. This population contains female plants having minute, nonexserted anthers and male plants having vestigial stigmas. Instead of having the normal glabrous lemmas, some of the female plants are pubescent. A second population is found in a disturbed area above the Timberline garden and contains plants which differ from the typical forms in having large, though aborted, anthers. Here also the ligules, which in the plants of the Sierra Nevada are typically 1 mm long, are up to and slightly above 3 mm long. Poa nervosa here occurs together with plants of the P. Canbyi-epilis group, which have normal anthers and a ligule up to 4 mm long. The chromosome number typical of most plants of P. nervosa of the central Sierra Nevada is 2n = 63, but the one plant of this variant population which has been counted has 2n = ca. 80. Another population, found at Tuolumne Meadows in the Sierra Nevada, also contains plants that grow intermingled with Poas of the Canbyi-epilis complex. The anthers of plants of this population are extremely small, but the unusual ligule length of about 3 mm again occurs. The presence of aborted anthers and of longer ligules in

these last two populations suggests that for some of its variations *Poa nervosa* may depend upon sexual reproduction using genes carried by the pollen of other species of *Poa*.

POA CYTOLOGY

PAUL GRUN AND EDWARD L. TRIPLETT

Chromosome pairing in Poa hybrids. As described in Year Book No. 50 (1950-1951, pp. 111-112), chromosomes of F_1 plants of intersectional bluegrass hybrids frequently pair as regularly as do the chromosomes of well established bluegrass species. It might then appear that no cytological barrier exists to the free exchange and recombination of genes of such species. Study of the F2 progeny of such a hybrid has, however, shown that at least a partial cytological barrier exists between the species. The female parent of this hybrid is Poa scabrella, Las Posas (4212-3), and the male parent is P. pratensis, Mather (4253-4). Their chromosome numbers are 2n = 84 (12 sets) and 2n = ca. 68 (ca. 10 sets), respectively. The parent species represent morphologically contrasting sections of the genus. The P. scabrella parent is a winter-active, summer-dormant plant from the southern California Coast Ranges, and the pratensis parent a winter-dormant but summer-active grass from 4500 feet altitude in the central Sierra Nevada. They also differ in at least five morphological characters. The F1 hybrid (4557-10) is intermediate in several of its morphological characters and has an intermediate number of chromosomes, namely, 2n = ca. 75. It is winter-active and partially summer-active.

Meiosis is reasonably regular in both parents and the hybrid, the *Poa scabrella* parent having 1.3 ± 0.7 univalents, the *P. pratensis* parent 3.6 ± 0.9 univalents, and the F_1 3.1 ± 0.9 univalents. In each case the standard errors used are based on counts of 20 metaphase plates and indicate

the possible variation of the mean to be expected in 99 out of 100 trials. In calculating the standard errors of these data the fact had to be taken into account that the curves were predominantly Poisson distributions.

The F₁ was vigorous and sexual, and after self-pollination at Stanford it produced an F2 that was variable in vigor and on an average weaker than the parents and the F₁. This particular F₂ was, nevertheless, one of the most vigorous of the sexual interspecific hybrids of Poa. Study of chromosome pairing in 21 plants of this F2 revealed that partial barriers to chromosome interchange had arisen. One individual was found with relatively normal pairing, having 3.4±1.0 univalents; but other plants differed through a continuous series of intermediates ranging to the opposite extreme, a plant with 23.7 ± 3.8 univalents. Of the 21 F2 plants, only one was as regular as the parents and the F1 hybrid, the other 20 having significantly more univalents.

Several attempts were made to correlate morphological characters and the chromosome pairing in the F2. Since there was variation in the vigor of the F2, the correlation between the number of stems produced on a plant (a measure of vigor) and its frequency of univalents was calculated and found to be very close to o. Within this F2 there was a limited segregation for the morphological characters that distinguish the parents. The F2 individuals were therefore classified for those characters which showed segregation in relation to their degree of scabrella characteristics. The scabrella character was coded as I, and its corresponding pratensis equivalent as 9. The characters used included amount of lemma pubescence, date of first flowering, number of florets per spikelet, and length of ligule. There was no noticeable correlation between any of these characters and the frequency of univalents. Since the parents and the intermediate F_1 were regular, the possibility was considered that only plants midway between the F_1 and either parent would be irregular. To test all characters taken together, their code values were added into an index. These index values, also, were not correlated with the frequency of univalents, plants having the same index as the F_1 being, in some cases, very much more irregular than was the F_1 .

Chromosome numbers in Poa hybrids. In addition to the earlier counts of chromosome numbers of Poa hybrids made by Marguerite Hartung, reported in Year Book No. 43 (1943–1944, p. 76), counts have been made during the past few years by Paul Grun, E. L. Triplett, I. P. Crawford, and Mary Wagner. The chromosome numbers found for the hybrids fall into two classes, one of which reflects the fertilization of a reduced, and the other the fertilization of an unreduced egg.

Chromosome counts of 93 hybrids representing 28 different combinations of intersectional *Poa* hybrids have been accumulated and are listed in table 3. The chromosome numbers of the hybrids indicate that both reduced and unreduced eggs have been fertilized in producing hybrids. In the few cases in which only reduced or unreduced eggs have been found, too few hybrids have been investigated to justify the conclusion that only one type of hybrid could be formed.

The difference in chromosome number between a hybrid derived from fertilization of a reduced and one from an unreduced egg is large. In the hybrid Poa ampla, Albion, Washington, $2n=64 \times P$. pratensis, Mather, California, 2n=68, the F_1 's produced from unreduced eggs had approximately 98 chromosomes, whereas those from reduced eggs had about 66 chromosomes. Of the 98 chromosomes in

F₁'s derived from the fertilization of an unreduced egg, 64 came from ampla and the remaining 34 from pratensis; the 66chromosome F₁ from a reduced egg contained approximately 32 ampla chromosomes and 34 pratensis chromosomes. Superimposed upon this large difference there is a smaller range of variation in chromosome numbers within each class. Thus, in the class of hybrids from unreduced eggs of the above cross, the somatic chromosome number ranged from 92 to over 100. This variation represents chromosomes gained or lost because of irregularity of meiotic divisions (Year Book No. 50, 1950-1951, pp. 111-112).

The differences in chromosome number between hybrid products of reduced and unreduced eggs help to explain some of the morphological differences between F_1 individuals. This difference can best be assayed in the case of individual crosses in which both types of hybrid are produced by the same parents.

The Poa ampla, Albion \times P. pratensis, Mather combination has, as was stated above, produced hybrids of both types. Some of the characteristics of these plants are summarized in table 4. The character glaucousness is graded so that the figure 9 indicates a very blue leaf color, particularly in midsummer, and I a dark green leaf lacking glaucousness. Similarly, the figure o describing leaf stiffness indicates a very stiff leaf, and I a limp one. The F₁ plants 4535-4 and -52 arose through the fertilization of a reduced egg of ampla and are very much less ampla-like than are the individuals 4535-2 and -6, F1's derived from fertilization of an unreduced ampla egg. In the F_1 of P. ampla, Albion \times P. pratensis. Athabasca, there are similarly two chromosomal forms, one of the reduced, the other of the unreduced ovule type, and these are also correlated with morphological differences of the same sort, the differ-

TABLE 3
Chromosome numbers of Poa Hybrids

Parents		2n chromosome numbers			
	<i>ਹ</i>	Parents			
Q		\$ q,		F1 hybrids	
ampla, Albion	× pratensis, Mather	64	68	63, 70, 92, 93, 94, 96 98, 100, 104	
ampla, Kahlotus	× pratensis, Athabasca	64	70	77, 79, ca. 83, 84	
ampla, Heise	× pratensis, Newport	70	81	73, 75, ca. 79	
ampla, Condon	× pratensis, unknown	63		ca. 98	
ampla, Enterprise	× pratensis, unknown	63		96	
ampla, Albion	× pratensis, Lapland	63	74	63, 68, 70, 72, 73	
ımpla, Albion	× pratensis, Athabasca	64	70	75, 103	
cabrella, Watsonville	× pratensis, Athabasca	82	70	71, ca. 119	
cabrella, Las Posas	× pratensis, Lapland	82-84	74	76, 79, ca. 110	
scabrella, Las Posas	× pratensis, Mather	82-84	68	64, 67, 68, 70, 71, 74, 7	
scabrella, Las Posas	× pratensis, Athabasca	84	70	69, 70, 71, 72, 73, ca. 7	
scabrella, Las Posas	× ampla, Kahlotus	82-84	63-64	75–76	
cabrella, Las Posas	× compressa, Chorsum	84	50	65, 105, ca. 115	
cabrella, Las Posas	× compressa, Crescent Mills	84	42	63-65, 101	
ampla, Albion	× compressa, Chorsum	64	50	ca. 50, ca. 62, 87	
compressa, Chorsum	× ampla, Albion	50	64	56, 59	
impla, Spokane	× compressa, Crescent Mills	63	42	ca. 81, 84	
ampla, Albion	× arida, N. Platte		63	ca. 50, 59, 61	
<i>irida</i> , N. Platte	\times ampla, Albion	63		ca. 97, 98	
scabrella, Las Posas	× arida, N. Platte	84	63	67	
arida, N. Platte	× scabrella, Las Posas	63	84	ca. 105	
Canbyi, Blue Mts.	× pratensis, Athabasca	84	70	77	
gracillima, Slate Creek gracillima, Fallen Leaf	× scabrella, Yager Creek	84	84	86	
Lake	× pratensis, Athabasca	86	70	ca. 72, ca. 74, ca. 76	
scabrella, Lucerne	× arachnifera, Stillwater	ca. 63	58	57	

TABLE 4 $\text{Morphology of four Poa ampla} \times \text{P. pratensis hybrids as related to the type of egg fertilized }$

Species or hybrid	2 <i>n</i>	Glaucous- ness, grade (1-9)	Leaf stiffness, grade (1–9)	Leaf width (mm)	Leaf form
P ₁ : Poa ampla, Albion	64	9	7	5	Flat
P ₂ : Poa pratensis, Mather		1	2	3	Folded
4535-4 from reduced egg	63	2	2	3	Folded
4535-52 from reduced egg	70	2	2	3	Folded
453,5-2 from unreduced egg	96	9	8	5	Flat
4535-6 from unreduced egg	ca. 96	9	8	4	Flat

ence in glaucousness again being particularly striking.

Poa scabrella, Las Posas, California, when crossed with P. arida, North Platte, Nebraska, also produced both types of hybrid: one plant having scabrella as the female parent was the product of reduced sex cells of both parents, and another with arida as the female parent originated from an unreduced egg of arida. The latter was more like arida, particularly in its summer activity, glaucousness, and rhizomes, than was its reciprocal counterpart produced from a reduced gamete of arida.

The combination $Poa\ ampla$, Albion \times $P.\ arida$, North Platte produced plants from a reduced egg when ampla was the female parent, but from an unreduced arida egg in the reciprocal hybrid. In this case also the $arida \times ampla F_1$, which had the two sets of arida chromosomes, was closer to $P.\ arida$ morphologically, particularly in rhizomes and leaf and stem lengths, than was the $ampla \times arida$ hybrid, which had one set of arida chromosomes.

These facts emphasize again the evolutionary flexibility of members of the genus *Poa*. Not only can widely unrelated species be crossed to form vigorous, fertile, constantly reproducing hybrids, but each cross can produce several different types of such hybrids. Such flexibility may go far in explaining the apparent evolutionary success of certain members of the genus.

GENETIC STRUCTURE OF CLIMATIC RACES AND SPECIES OF ACHILLEA

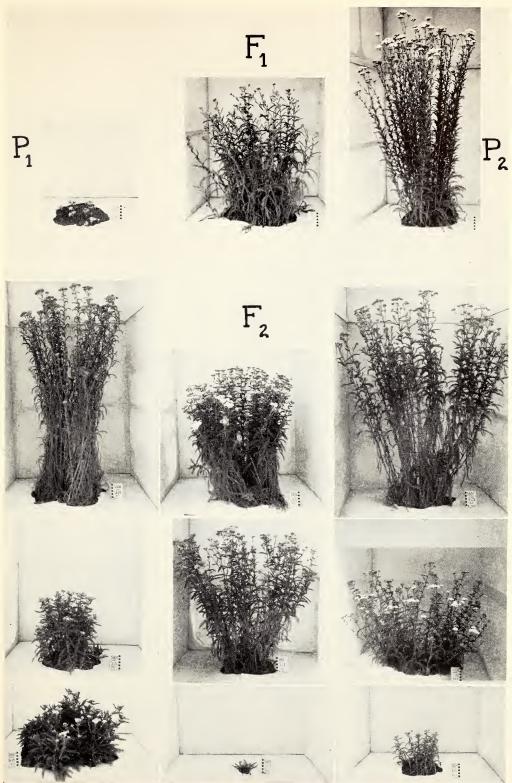
WILLIAM M. HIESEY AND MALCOLM A. NOBS

Most of the investigations in genetics made during the past half century have been concerned with relatively simple genetic systems, and have used characters whose inheritance could be related to the action of one, two, or three genes. In contrast, the hereditary composition of a climatic race or species is complex and is governed by hundreds or possibly thousands of genes expressed in recombinations making possible literally billions of different kinds of individual variants.

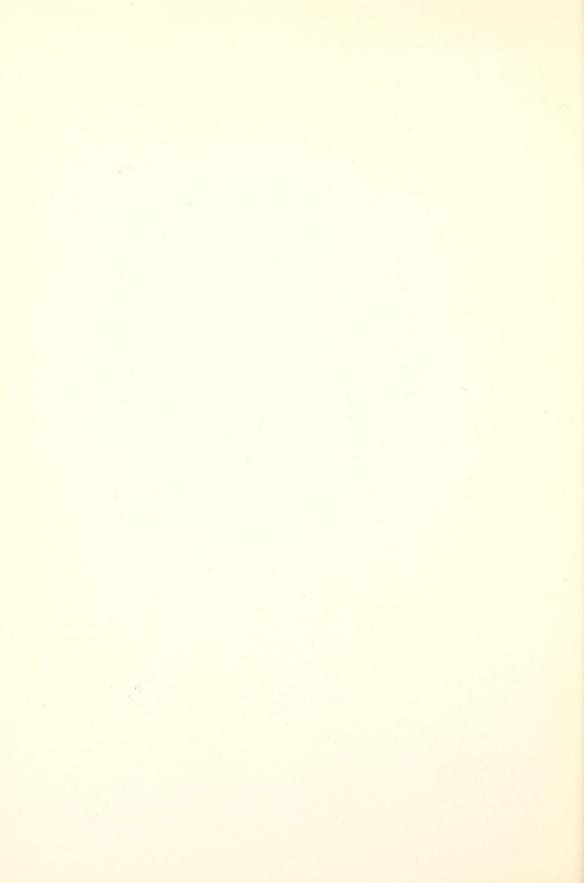
An analysis now completed of the genetic structure of contrasting altitudinal races of *Potentilla glandulosa* was briefly reviewed in Year Book No. 47 (1947–1948, pp. 106–110). A complementary study of the genetic characteristics of latitudinal races of the same and also of different species of the *Achillea millefolium* complex was outlined in the annual report of last year. The significance of the findings in *Achillea* to our understanding of variation and evolution in higher plants makes timely the following review.

Plate 1 illustrates a key experiment in crossing two forms of Achillea. In the topmost row on the left is shown a dwarf race of Achillea borealis originally from Kiska Island in the Aleutian chain, and on the right a giant form of the same species originally from the San Joaquin Valley in California. Between them is their F₁ hybrid. In the lower three rows is shown a sample of 9 plants of second-generation or F₂ progeny obtained from the hybrid. These progeny were all grown at the same time in the same field at Stanford, and those illustrated include the extreme recombination types as well as examples of the more numerous intermediate-type plants found in the population of some 1000 individuals.

Among the F₂ progeny are individuals that closely resemble in form many of the great host of variable forms of the species that are found in the wild in the many different kinds of environment in which it grows. Detailed studies on morphological characters distinguishing the parent forms, including length of stems, number of internodes, length of leaves, pubescence,



Parents, hybrid, and second-generation progeny of a cross between contrasting climatic races of *Achillea borealis*



habit of growth, mode of branching, and anthocyanin pigmentation, show that these characters are recombined in the F₂ in all possible ways. Furthermore, among the approximately 1000 F₂ progeny grown in the Stanford garden not a single plant resembles either parent in all its characteristics. With respect to a single character, however, usually several individuals can be found which match one parent or the other, and from the frequency of such types some estimate of the number of genes governing that particular character can be made.

Even more striking and more significant from an evolutionary point of view than the recombination of morphological characters is the shuffling of physiological characters in the F2 progeny, as expressed by differences in seasonal periodicity, in rate of development, and in capacity to survive under the arid climatic conditions at Stanford. The Kiska parent when grown at Stanford is winter-dormant, develops flowering stems rapidly in spring, and tends to die during the summer for lack of sufficient water. The Selma parent, by contrast, is winter-active, much slower in the development of flowering stems, and relatively much more tolerant to drought, although it itself is a mesophytic race that grows naturally in areas with ample underground water. In the F2 progeny a complete scrambling of characters, both morphological and physiological, seems to have taken place.

In order to determine whether significant correlations can be found between morphological and physiological characteristics, and also whether these can be related to the capacity of given types to survive in given environments, 300 of the segregating F₂ individuals, along with the parental forms, have been cloned and planted at the Stanford, Mather, and Timberline transplant stations this year. Each indi-

vidual clone will be studied during the next few years to determine its capacity to survive and develop in these three contrasting environments. This study of these progeny originally from contrasting latitudinal races will be of special interest in connection with the selection experiment on progeny from different altitudinal races of *Potentilla glandulosa* referred to above.

Recombinations similar in character to those obtained in the second-generation progeny from the cross between the dwarf Kiska and the tall San Joaquin Valley forms have been found in other crosses between climatic races both of Achillea borealis, having 2n = 54 chromosomes, and of A. lanulosa, having 2n = 36 chromosomes. In each hybrid progeny a similar result has been obtained, including the production of vigorous and fertile F1 and F2 generations and a complex recombination of parental characters in the second generation. The total range of segregation in the F₂ progenies differs, however, according to the extent of the differences between the original parents. Moreover, segregation products from crosses in the two levels of ploidy overlap widely in both physiological and morphological characters.

When, however, A. borealis from California is crossed with A. millefolium from Europe, both having the same number of chromosomes, vigorous F1 hybrids are produced but these are relatively sterile and yield few progeny. In one F2 population obtained by crossing a typical form of A. millefolium from Iceland and the giant race of A. borealis from the San Ioaquin Valley, the surviving progeny tended to aggregate around the parental types rather than to segregate freely as in the intraspecific crosses described above. The relative sterility of F_1 hybrids between A. millefolium and A. borealis is correlated with irregular chromosome pairing at meiosis, as described below by Ehrendorfer. Thus, although many forms of A. borealis from North America resemble A. mille-folium morphologically, they have diverged so far that their chromosome complements can no longer work in harmonious balance.

CYTOLOGY OF ACHILLEA HYBRIDS FRIEDRICH EHRENDORFER

Successful hybridization of different races of the Achillea millefolium complex was first reported in Year Book No. 49 (1949-1950, p. 107). Further data are given in Year Books Nos. 50 and 51. Only hybrids between races on the tetraploid and hexaploid chromosome level, respectively, have so far been obtained. It was thought that a cytological analysis of these hybrids would clarify our understanding of the relationships between the races involved and throw some light on the mechanisms of internal crossing barriers between different entities on the same chromosome level. This preliminary report gives an account of the meiosis in pollen mother cells of some of the parents, hybrids, and F2 offspring.

The meiotic behavior of the parental races is usually very regular even though they are tetraploid (n=18) and hexaploid (n=27). There are usually not more than 2 to 5 per cent of first metaphase plates with irregularities such as uni- and multivalents. Consequently, lagging univalents and fragments in anaphase stages and micronuclei in telophase are very rare or missing in material from the wild. Exceptional plants, however, have higher uni- and multivalent frequencies, more telophase irregularities, and higher frequencies of sterile pollen grains. In addition to chromosomal behavior, however, physiological factors are of utmost importance in determining the final quality of pollen. Irregularities that are definitely not chromosomal sometimes lead to improper functioning of the spindle or interfere in other stages of development. Interpretation of pollen quality in terms of chromosomal behavior alone, therefore, is not reliable.

On the tetraploid chromosome level, four hybrids between different races of the North American Achillea lanulosa were studied. These races are described in some detail in a later section of this report. Two hybrids, one between a race from the redwood region and a coastal type from southwestern Oregon (plants 5133-112, -113), and the other between the xeric subspecies lanulosa and the Oregon coastal form (5134-2, -4), are normal. Hybrids between the same coastal race and a form of subspecies alpicola from Big Horn Lake in the high Sierra Nevada (5801-102), and between the coastal race and a form from near Vera Cruz, southeastern Mexico (5132-1), show a number of disturbances. In the hybrid with alpicola there are lagging chromosomes, uni- and multivalents, and bridges and fragments, with the result that approximately 25 per cent of the tetrads have micronuclei. The hybrid with Vera Cruz is even more irregular, showing similar disturbances in approximately 50 per cent of the tetrads and occasionally a total breakdown of first or second anaphase and the formation of restitution nuclei.

On the hexaploid chromosome level, hybrids were obtained between different races of the North American Achillea borealis. A cross between the giant California race from the San Joaquin Valley and a form of the coastal subspecies arenicola is normal (5129-118), but hybrids between the same giant race and both dune and alpine forms of subspecies borealis from Kiska Island in the Aleutian chain show slight irregularities which are definitely chromosomal; these are also found in the reciprocals (5121-118, 5125-110, 5126-115) of these same combinations. In these there are lagging

chromosomes and uni- and multivalents, but no restitution nuclei. The frequency of micronuclei in tetrads ranges in these different hybrids from approximately 5 to 15 per cent. Here, as in the other crosses in which the chromosomal behavior of the parental generation has been checked, no irregularities could be detected in the parents.

Also on the hexaploid level, hybrids between Achillea millefolium from Lapland and from Iceland in northern Europe and the giant race from California were studied (5130-1 and 5131-1). In contrast with the hybrids within A. lanulosa and A. borealis, which are normal or only somewhat irregular, strong irregularities are encountered in the meiosis of the pollen mother cells of these interspecific hybrids. In addition to the common occurrence of lagging chromosomes, uni- and multivalents, bridges and fragments, and restitution nuclei, there is a general physiological lability of the meiotic processes. This lability expresses itself in a strong variability in the degree of meiotic breakdown as seen in different anthers and even in different pollen sacs of the same flower, and ranges from slight disturbances to a total breakdown of meiosis and abortion of the contents of entire pollen sacs.

The cytological irregularities in the hybrids Achillea millefolium × borealis are accompanied by a relatively high degree of seed sterility and a low viability of the F₂ offspring. It is of interest that there is some difference in fertility between the combinations Iceland × California giant, which is relatively the more regular, and Lapland × the same California giant race. In the former cross an analysis of eight F₂ plants has been made to determine whether irregular meiosis in the first hybrid generation leads to varying chromosome number and higher polyploidy in the second generation. This was found not to be the

case, for none of the plants deviated significantly from the original number, n = 27. These F_2 plants, however, were highly diverse with respect to the degree of their meiotic irregularities; they ranged from almost normal to strongly irregular, that is, more irregular than the F_1 . Apparently there is segregation in the F_2 progeny in their degree of meiotic disturbance.

The cytological data reviewed above are in line with the interpretation of the relations of the different races of the Achillea millefolium complex set forth in the following section. These data indicate that in Achillea there is a gradual development of internal crossing barriers between races and species on the same chromosome level. This is apparently due to chromosome repatterning, resulting in decreased homology and upset pairing balance. Some of the hybrids show multi- and univalents, bridges and fragments, and subsequent formation of micronuclei or even of restitution nuclei. This tendency seems to be accentuated in some instances by physiological lability of the meiotic processes.

CYTOTAXONOMIC STUDIES IN ACHILLEA FRIEDRICH EHRENDORFER

Previous studies on the northern circumpolar Achillea millefolium complex were mainly confined to races in western North America. Clausen, Keck, and Hiesey (Carnegie Pubs. 520 and 581) showed the existence of an amazing array of physiologically distinct tetraploid and hexaploid ecotypes which occur from southern California to Alaska, and from the Pacific Ocean through the Coast Ranges, the marshes of the San Joaquin River, and the foothills up to above tree line in the Sierra Nevada. W. E. Lawrence (Amer. Jour. Bot., vol. 34, pp. 538-545, 1947) continued cytological investigations on the group and outlined the general distribution of the tetraploid and hexaploid races in the Pacific states and Alaska.

Preliminary unpublished work by Ehrendorfer around Vienna in central Europe revealed the existence of an even greater differentiation of races and species. There two very different diploid species (n=9)were discovered: Achillea setacea W. & K., occurring on very dry sand steppes, and A. asplenifolia Vent., a member of the low moor vegetation (Molinietum). Furthermore, A. tomentosa L., a species which does not seem to belong to the A. millefolium complex, was also found to be diploid. A tetraploid chromosome number (n=18) is characteristic for A. collina Becker, which occurs in dry grasslands and open oak forests. Achillea pannonica Scheele, which is morphologically very close to A. colling and can be found on very dry slopes, seems generally to be octoploid (n=36). Finally, A. millefolium L., sensu stricto, is a hexaploid species (n =27) confined to plant communities where there is a good water supply during the summer; the center of distribution of this species in Europe is more northern. Achillea stricta Schleicher occurs in open mountain forests and is also hexaploid. It belongs to the A. tanacetifolia All. complex, which is not sharply separated from the A. millefolium group.

This interesting and highly complex situation in Europe made it desirable to continue and widen the studies on the New World forms of the *Achillea millefolium* group. A thorough knowledge of the distribution of the various chromosome races and their morphological and ecological similarities and differences seemed to offer the best basis for further studies toward the final goal of understanding the development of the racial complex in time and space.

Determination of the chromosome numbers of more than 70 new strains of the

Achillea millefolium complex brought the total of known strains in North America to more than 170. Considerable interest was centered upon the question whether it would be possible to distinguish between tetraploids and hexaploids on the basis of herbarium material. The finding that a correlation exists between pollen size and chromosome number made such a distinction possible. The measurement used was maximal inner width of good pollen grains taken from recently opened flowers that were boiled in acetocarmine. Pollen diameters in tetraploids range between 20.5 and 23.5 microns, and in hexaploids between 24 and 28 microns. European diploids fit in this scale, for their pollen grains are below 20 microns, but it still remains to be seen whether the scale also applies to higher polyploid forms in Europe.

The addition of new counts of chromosome numbers and the wide use of the correlation between chromosome number and pollen diameter for the determination of the polyploid level of herbarium specimens from areas without or with insufficient cytological data now provide a fairly complete picture of the distribution of tetraploid and hexaploid Achilleas in the western United States. The following sketch of the racial structure of the Achillea millefolium complex in North America is based on these data, on the results from crossing experiments, on careful morphological studies of the rich collection of living plants of the Carnegie Institution's Department of Plant Biology, and on extensive herbarium collections. A consideration of the ecological and geographical features of the various populations was an essential part of this analysis.

As an introduction to a discussion of the numerous races of the *Achillea millefolium* group, it seems worth while to consider some of their common features. This will help in understanding the many obstacles which have blocked and continue partially to block an understanding and satisfying classification of that huge complex of forms which have established themselves very successfully in an amazingly large number of plant communities all over the northern hemisphere.

Natural local populations of Achillea usually contain a wealth of different genotypes. Nothing is more informative in this respect—and at the same time more baffling for the taxonomist—than to see growing under uniform conditions series of variable plants that originated from single mother plants in the wild. The genetic variability displayed by such test cultures likewise affects morphological features such as height of stem, number of nodes, form and size of leaves, and degree of leaf cut, and such ecological-physiological characters as time of flowering and periodicity. natural populations these characters can easily become recombined, because the Achilleas are mainly cross-fertilizers. Vegetative propagation is limited to varying amounts of development of runners, and no apomixis is known to occur in the group. Achilleas are sturdy and can stand rough treatment; they can adapt themselves to a considerable range of conditions and become strongly modified at the same time. These characteristics certainly are in a large degree responsible for the wide distribution and the aggressive and often weedy character of many races of the Achillea millefolium complex. Because of the more or less continuous distribution and the wide ecological amplitude, geographic and ecologic barriers between the segments of the complex have never been very effective in isolating sharply defined entities. With the exception of the establishment of different levels of polyploidy, the evolution of internal crossing barriers has also obviously been rather lacking, as is demonstrated by the

ability of extremely different forms within the tetraploid and the hexaploid chromosome levels to intercross successfully.

These general features of the races of Achillea millefolium—high local variability, little consistent morphological differentiation over larger areas, many modifications, and few effective barriers to wide and often introgressive gene flow-account for the many difficulties which bar the way to a satisfying taxonomic classification and make the complex a real "crux botanicorum." With these difficulties in mind, it becomes clear why our concept of racial entities in Achillea does not and cannot represent more than groups of populations which are close to one another with respect to their cytological, morphological, ecological, and geographical features, but which are nearly always connected by intermediate populations whose correct position remains obscure or can be described only in terms of statistical frequencies.

The races representing the Achillea millefolium complex in North America are almost exclusively tetra- and hexaploid. Various chromosomal races replace one another in different localities and are only very rarely found growing side by side. Whereas the tetraploid forms range in a variety of different ecotypes from the coast of the Pacific to the northeastern United States, with maximum development in the mountainous areas of the West, the hexaploids are entirely confined to the Pacific coast. Where tetra- and hexaploid races occur in similar habitats, parallel ecological entities have evolved. In spite of very careful morphological analysis, it has so far been impossible to find good morphological characters except pollen size which would permit separation of the two chromosome levels as a whole. With the present lack of information on the closely related forms in eastern Asia, it seems premature to decide definitely on the taxonomic status of the two North American race complexes. Tentatively, therefore, the specific names *Achillea lanulosa* Nutt. and *A. borealis* Bong. are here retained to designate the tetraploid and hexaploid forms, respectively, as they occur in North America.

The tetraploids. In discussing the tetraploid forms it seems proper to deal first with some peculiar races which occur as small islands or larger bulges between the hexaploids on the Pacific coast and which obviously have played an important role in the evolution of the latter. With the exception of a race on the Olympic Peninsula, these races are all characterized by tall or stout stems, many condensed internodes, strongly divided, broad, and definitely auricled leaves, and lack of strong pubescence on the stem and on the leaves. In the habitats where they occur they have ample water supply all through the year and are both summer- and winter-active. Among these coastal tetraploids there is, first, a race of the marshlands of Marin, Napa, Solano, and Contra Costa Counties in California which has been named A. puberula Rydbg. It is very tall and has firm, somewhat leathery and spinulose leaves.

Another tetraploid race occurs in openings and at the borders of otherwise dense redwood or similar coniferous forests in Humboldt and Del Norte Counties of northern California and in Curry and Coos Counties of southwestern Oregon. It is a tall but usually tender plant with very broad leaves which are deeply cut into fine linear segments. In its southern area, toward the coast and on the drier slopes of the Coast Ranges, this characteristic race of the relict Sequoia sempervirens forest is surrounded by hexaploids. Farther north, the redwood race of Achillea is intimately connected with the third of the tetraploid races on the Pacific coast, a coastal ecotype ranging along exposed places from Brookings to Bandon in Curry and Coos Counties of southwestern Oregon. It is a lower but stout plant with somewhat thickish leaves which are densely divided into short linear segments. The two latter tetraploid races, namely the redwood and coastal forms, and another tetraploid race, to be mentioned later, which advances westward from drier coniferous mountain forests farther east, produce a solid tetraploid barrier extending at least 120 miles from north to south, which separates the California hexaploids from those in Oregon and Washington.

The hexaploid group of Achillea in Oregon and Washington is again separated from the northernmost representatives of the hexaploid A. borealis in coastal Alaska, the Aleutian Islands, and northern British Columbia by populations of tetraploid A. lanulosa which reach from the interior out onto the Pacific coast of the Olympic Peninsula in Jefferson and Clallam Counties, Washington, and to Vancouver Island, British Columbia. This northern coastal tetraploid race shows a combination of the morphological characters of the redwood and the coastal races of southwestern Oregon and of strongly hairy forms from the interior which come near the coast in dry grasslands of the "prairies" of the Puget Sound area and even occur on dry granitic outcrops on the leeward side of the islands in the Sound. The influence of these interior forms is shown principally in a strong appressed white pubescence on the stem, mainly under the leaf bases, a pubescence which gives the stem a somewhat shaggy appearance. Habit and leaf cut of these forms are either more like those of the redwood or like those of the coastal race of southwestern Oregon, depending on the habitat and thereby expressing a certain ecotypic differentiation. There are numerous forms intermediate toward the pubescent interior race, mainly on the east side of the Olympic Peninsula.

The tetraploid Achilleas in the more interior mountain areas of the Sierra Nevada and the Cascades to the Rocky Mountains, and from Mexico up to central Alaska, form another group of about three more closely related races. They are on the whole smaller plants, often with long runners and usually with fewer internodes; their stems and leaves are beset with scattered or dense long hairs; their leaves are less divided, having segments that often are broader and more contracted; the pedicels supporting the heads are mostly densely covered with short curled and longer straight hairs. These tetraploid montane races are often exposed to a period of drought during the summer and are mainly winter-dormant, being covered during that time by snow. The most xerophytic of the three taxonomically discernible entities of Achillea lanulosa in the western mountain states is the typical form of that species, subspecies lanulosa. It is a short and very woolly plant with relatively many internodes, narrow leaves, a contracted inflorescence, and straw-colored bracts. It is found in dry grassland, prairies, and the interior sagebrush steppes, and may extend into open and dry Pinus ponderosa forests. It ranges from southern British Columbia through Washington, Oregon, and Idaho to Nevada, Utah, and Colorado, and in scattered places down to northern Mexico. Very polymorphic is a second type found in drier coniferous forest, which is characterized by broader, deeper-cut greenish leaves and more open inflorescences. This type often has only long, thin runners; it occurs typically in open, moderately dry places on stony outcroppings and patches of grassland in the zones of mixed coniferous mountain forests from central Alaska south into northern Mexico and from the Sierras, the Siskiyou Mountains, and the Cascades to the Rocky Mountains, and may extend into northeastern Canada. In respect to ecology and morphology this race more or less connects the redwood type, the xerophilous subspecies lanulosa, and a group of alpine forms of A. lanulosa in western North America which has been called subspecies alpicola (Rydbg.) Keck. This alpine and subalpine racial complex is represented by small plants usually not surpassing 20 cm in height. They have inflorescences with relatively few but large heads, and bracts mostly with dark brown margins; their leaf shape and pubescence are rather variable. Forms with broader greenish leaves are common in the northern parts of the distribution of alpicola. These northern forms of *alpicola* are close to the type from montane coniferous forests, whereas in the southern parts of the distribution only strongly hairy, gray forms with contracted and much smaller leaves have been observed, which are close to subspecies lanulosa.

In the middle western states and south through Texas to southeastern Mexico, another tetraploid race is encountered which, together with forms in the northeastern states, forms a third group of more closely related racial entities within A. lanulosa. Both the middle western and the northeastern races have tall stems with many internodes, deeply and divaricately divided leaves that are broad and auricled, open inflorescences, short curly hairs and glands scattered on the pedicels of the heads, and straw-colored bracts. These characters and the lack of long woolly pubescence remind one of the western redwood type. The presence, however, of numerous short hairs and stalked glands on the middle stem leaves in the forms from the Middle West, and the dark green, bluntly divided leaves of the northeastern forms, which are similar to the European A. millefolium, permit a taxonomic distinction. Information on the ecology and physiology of these races is still lacking. The forms of the Middle West are obviously members of the prairie vegetation, whereas the eastern forms are mainly found in openings and rocky outcroppings in the area of the mixed deciduous forest. Toward the north into the Canadian plains, both racial entities seem to grade into forms which are similar to the racial complex of the western coniferous mountain forest, but the specimens seen from these areas are too few to permit a full interpretation.

The hexaploids. The hexaploid races of the Achillea millefolium complex in North America, called A. borealis Bong., are confined to the west coast and are split up into three isolated units which are geographically separated from one another by the intervening tetraploid populations described above.

The southern group of A. borealis in California ranges from the Channel Islands along the Pacific coast through the Coast Ranges, the upper San Joaquin River basin, and northward to Trinity, Humboldt, and Del Norte Counties. The most widely distributed entity is subspecies californica (Pollard) Keck, a plant of open and dry oak forests and parklands, sometimes with admixed pines. It is somewhat similar to the tetraploid redwood race, but the plants are usually stouter; there is always a well developed pubescence of long hairs on stems and leaves, and the flower heads are larger. Subspecies californica is summer- and winter-active in the moister areas, but summer-dormant and usually more hairy in the drier interior, thus showing distinct ecological differentiation. Along the coast a maritime ecotype, subspecies arenicola (Heller) Keck, has been This race is shorter and established. stouter than californica and has crowded, often very hairy leaves which are very

intricately divided into lanceolate-ovate, divaricate segments. Similarities to the tetraploid maritime ecotype from southwestern Oregon are obvious. In the marshes of the upper San Joaquin Valley a gigantic summer- and winter-active race of A. borealis occurs, which has been called A. gigantea Pollard. This race is mostly strongly white-hairy and has a very contracted base with a strong vertical root and very short vertical runners which form a dense rootstock. Its ecology and habit indicate relations to the tetraploid A. puberula marsh type.

The Oregon-Washington group of hexaploids contains maritime forms, mainly on sand dunes, which extend from North Bend, Oregon north to Copalis Beach, Washington, and shade forms of the Coast Range forests between the Pacific and the Cascades south of the Olympic Peninsula in Washington. The maritime forms are scarcely different in morphology from the tetraploid maritime types on the Olympic Peninsula, but are rather distinct from the California *A. borealis* ssp. *arenicola*; the Coast Range forest forms are very close to *A. borealis* ssp. *californica*.

The northernmost racial complex of A. borealis, subspecies borealis, is a morphologically relatively distinct entity characterized by short stems, and by broad and finely divided green leaves, of which the upper ones are involucral and subtend the inflorescence; the pedicels of the relatively few flower heads are densely covered with long, often yellowish hairs, and the bracts of the heads have more or less dark brown margins. Achillea borealis ssp. borealis is known to occur from Goose Island, north of Vancouver Island in British Columbia, along the Alaskan coast and the Aleutian Islands north to the shores of the Arctic Sea. It occurs in open arctic vegetation near the seacoast and is largely summerand winter-active. The tetraploid races of A. lanulosa that are most similar to A. borealis ssp. borealis are some of the northern forms of subspecies alpicola and also types from coniferous forests of the interior of Alaska.

In conclusion, some consideration may be given to a tentative interpretation of the recent racial structure of the Achillea millefolium complex in terms of evolutionary history of this group in North America. It is notable that, unlike the Achilleas in central Europe, those of North America have no known diploid forms. Whether the diploids have become extinct there or whether only tetraploids migrated into the New World remains uncertain, but the existence of tetraploid races in central and eastern Asia which are very similar to forms of A. lanulosa seems to support the latter hypothesis. Furthermore, the lack of crossing barriers between extreme forms within the tetraploid or hexaploid chromosome level is a fact of considerable evolutionary significance, permitting secondary contacts and infiltrations between formerly separated entities. On the tetraploid chromosome level, the mesic relict races in marshes around San Francisco Bay, in the redwood forests of northern California and southeastern Oregon, and along the coast of southeastern Oregon probably represent segments of a relatively primitive racial entity, which may have existed in that area fairly unchanged for very long periods. The forms of the coniferous mountain forests are probably very old too, whereas the xeric subspecies lanulosa and the alpine subspecies alpicola occur only in definitely more recent environments. For the forms on the Olympic Peninsula a postglacial development through infiltration between mesic coastal and xeric interior forms is very likely. The history of the hexaploids offers some puzzles. The crossability of extreme forms is in favor of a common

origin of the three geographic segments of *A. borealis*. On the other hand, their lack of common characters differentiating them from the tetraploids, their affinities with adjoining tetraploids, and especially their wide and complete separation by intervening tetraploid races which are at least partially very old, favor the assumption of polyphyletic evolution.

As may be seen from the foregoing report, many questions still remain open regarding the racial structure and the possible evolution of the *Achillea millefolium* complex, but further studies mainly on the important races in Europe and Asia are certain to yield more valuable information on this group and on our general knowledge of the intricate laws which underlie the creation of new forms.

Comparative Physiology of Ecologic Races

WILLIAM M. HIESEY AND HAROLD W. MILNER

Basic information on the comparative physiology of contrasting ecologic races of the same or closely related species is essential to further understanding of the mechanisms of natural selection in different kinds of environment. This need has been emphasized repeatedly by questions raised during the experimental taxonomy investigations. Progress in this direction has been made in the exploratory studies on the growth and development of Achillea and Poa under controlled conditions at the California Institute of Technology, reported in previous Year Books (No. 45, 1945-1946, pp. 112-116; No. 50, 1950-1951, pp. 99-105). These studies show that distinct climatic races may have a very different growth response when subjected to the same combinations of day and night temperature.

From such experiments and the extensive transplant investigations carried out

at Stanford, Mather, and Timberline, it is apparent that distinct climatic races differ markedly in their internal functions, and that in the last analysis these differences govern the direction of natural selection. It is not known what these physiological differences are. During the current year, work has been started on a program designed to investigate the basic physiological characteristics of ecologically different races.

Present efforts focus on the development of suitable techniques for measuring the rates of respiration and of assimilation under various conditions of temperature, light intensity, and humidity. These metabolic processes, in the complex living systems exemplified by higher plants, are influenced by changes in any one of several environmental factors. The development of satisfactorily precise methods for variation of the environment and for measurement of the induced physiological response of the plant is the first essential step toward an attack on the broader problems of the investigation.

BIOCHEMICAL INVESTIGATIONS

Outdoor Growth of Chlorella with Controlled Temperature and Different Light Intensities

HAROLD W. MILNER

In last year's report the outdoor culture of *Chlorella* in a rocking tray was described. The use of this apparatus demonstrated the feasibility of growing *Chlorella* at a high cell density in a thin layer of culture exposed to direct sunlight. The best average yield obtained from the tray was 8.2 g dry weight of *Chlorella* per square meter per day over a period of three weeks. This yield is equivalent to 73 pounds of dry *Chlorella* per acre per day. By extrapolation from previous small-scale experiments in flasks it had been estimated that a yield of 220 pounds per acre per day might be possible in summer.

The difference between the actual and the expected yield from the rocking tray was attributed to lack of adequate control of temperature and light intensity. On hot days the tray had to be partially shaded to prevent overheating the culture. Both an unfavorably high culture temperature and a reduction of the quantity of light would tend to lower the yield of algae. In order to measure the effect of temperature and light intensity on *Chlorella* cultures grow-

ing outdoors, a number of cultures were grown simultaneously with different temperatures and light intensities.

Three thermostatically controlled water baths were used. The Chlorella cultures were contained in glass tubes about 1.3 cm in diameter inside. The tubes were bent so that when the ends were vertical the straight section between them was nearly horizontal. The gently sloping section of the tube held the culture and was placed just under the surface of the water in the bath. The culture tubes were set in a northsouth direction near the center of the bath in order to obtain the maximum amount of sunlight per day. Since neither the volume of culture nor the area of culture exposed to sunlight was the same for the different tubes, all the yields of Chorella were calculated on a unit area basis in order that they might be comparable. The yield from duplicate culture tubes in one experiment differed by less than 10 per cent. The relative yields at different temperatures in separate experiments also agreed within 10 per cent.

Inocula for the experimental cultures were taken from a *Chlorella* culture growing outdoors in a 5-gallon bottle. The cells were centrifuged from the parent culture

and were resuspended in fresh medium at the desired cell density. The composition of the medium was the same as that previously used in this laboratory for growing Chlorella in quantity (Year Book No. 43, 1943-1944, p. 66). The cultures in the tubes were stirred and aerated by bubbles of air containing 5 per cent carbon dioxide. In addition, the cultures were agitated by hand several times daily to insure against settling of the cells. At the end of the growth period under specified conditions, the culture was removed from the tube and its cell density was determined. The difference between the final density and that of the inoculum was the amount of growth, called the yield.

The yields from outdoor *Chlorella* cultures in different experiments were obtained under conditions of uncontrolled duration and intensity of illumination. Therefore control cultures were grown each time, illuminated with full sunlight and maintained continuously at 25° C. The yield from each experimental culture was expressed as the percentage of the yield from its control. Thus, the results of several experiments could be combined to give an evaluation of the effect of a range of temperature or degrees of shading on the yield of *Chlorella*.

Three inoculation densities, 0.425, 1.45, and 7.50 g dry weight per liter, were tested. At the start, cultures with 0.425 g/l allowed much of the sunlight to pass through, those with 1.45 g/l had approximately the right cell density to give complete light absorption, and the ones with 7.50 g/l were illuminated only part way through. The greatest yield of *Chlorella* was obtained from the cultures started at the intermediate inoculation density, which therefore was used in starting all the following experiments. It is of interest to note that the percentage increase in cell mass was roughly inversely proportional to the density of the

inoculum. In grams of cells harvested, however, the yields from the low- and high-density inocula respectively were 54 and 65 per cent of the yield from the intermediate inoculum.

When the culture temperature was maintained continuously at 15° or 35°, the yield was 31 or 22 per cent, respectively, of the yield at 25° continuously. At 30° continuously the yield was 63 per cent and at 20° continuously 95 per cent of the yield from the control.

Under the conditions obtaining in these outdoor cultures, Chlorella can grow only in the light. During the hours of darkness it uses up a portion of the organic matter formed in the light. The adverse effect of high continuous temperature on the yield could be due to increased respiration of Chlorella in the dark. If so, the yield should be increased by keeping the culture cool at night. Cultures were grown at 25°, 30°, and 35° in the sunlight, then were kept at 20° or lower at night. The change from day to night temperature was made by moving the culture tube from one bath to another at dusk. The reverse change was made at dawn.

Using the data for cultures started with 1.45 g/l and grown in the sunlight, we can construct two curves showing the effect of temperature on the yield of *Chlorella* (fig. 1). The ordinate shows the yield at different temperatures as percentage of the yield from the control at 25° continuously. The solid line shows the yields at various temperatures maintained continuously. The broken line shows the yield when the daytime temperature is that indicated on the abscissa, but the night temperature is 20° in each case.

It is clear from the figure that the beneficial effect of the 20° night becomes larger as the day temperature is made higher. With a 30° day and 20° night the yield was double that obtained at 30° continu-

ously. Also it was the highest yield obtained in this series of experiments, 133 per cent of the control. With a 35° day and 20° night the effect was still larger. The yield was three times that at 35° continuously, but not so great as the yield from the control.

There is indication that a night temperature of 15° would be more favorable to increased yield than 20°. With a 25° day and 15° night the yield was 124 per cent

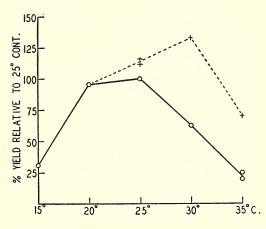


Fig. 1. Relation between temperature and yield from outdoor cultures of *Chlorella*. See text.

of the control, as compared with 113 per cent with a 25° day and 20° night. A still lower night temperature, 10°, gave no increase in yield over that of the control.

It is a matter of practical importance in the outdoor culture of *Chlorella* to know the effect on the yield caused by different degrees of illumination: sunlight, skylight, and shade or clouds. To compare the yield in skylight with that in sunlight, a vertical white board was placed so as to screen the culture tubes from direct sunlight, and at the same time to cut off only a small fraction of the skylight. The board was moved from time to time to compensate for the motion of its shadow relative to the cultures. The cultures grown in skylight gave 73 per cent the yield of those grown

in sunlight, both being maintained at the same temperature, 25°.

To test the effect of shade on the yield of *Chlorella*, a sheet of muslin was stretched over the water bath above the culture tubes. The light intensity on the cultures under the muslin was very roughly equivalent to the light they might have received on a cloudy day without the muslin. The yield from the shaded cultures was 55 per cent of that from the ones in sunlight, both at 25° continuous temperature.

The effect of a lowered night temperature was also tested on cultures grown in skylight and in shade. With shade, a 30° day, and a 15° night, the yield was 38 per cent higher than with shade and 30° continuously, in contrast with a 100 per cent increase in yield found for a 30° day and 20° night when the cultures were grown in sunlight. In either shade or skylight, with a 25° day and 15° night no increase in yield over 25° continuously was found, whereas in sunlight and with the same temperature conditions a 24 per cent increase in yield was noted. It is not clear why the increase in yield with a lower night than day temperature is smaller or absent when the intensity of the illumination during the day is less than full sun-

These experiments have merely scratched the surface of a fertile field for investigation. The data from such a small number of preliminary experiments do not have great quantitative significance. Rather, they indicate qualitatively the relations between yields of *Chlorella* under various sets of conditions. The quantitative magnitude of the differences in yield under the conditions tested could be stated only after much more exhaustive experimentation, accompanied by a considerable refinement of the method.

Yields of 14 to 18 g dry weight of Chlorella per square meter per day were

obtained in these experiments, made in November, when the cultures received only 8 to 9 hours of sunlight daily, but were growing at favorable temperatures and cell densities. The size and shape of the culture tubes were not, however, the most suitable for calculating yields per unit area, because of exposure to some light reflected from the white thermostats. Yields of about 25 g/m²/day might reasonably be expected in summer with 13 to 15 hours of sunlight per day. By inference, the 8.2 g/m²/day obtained in the rocking tray during the summer might be increased even as much as threefold if the tray were operated under optimum conditions of temperature, illumination, and cell density. This would correspond to a summer yield of 220 pounds per acre per day, in line with the figure expected by extrapolation from the small cultures referred to in the first paragraph.

QUANTITATIVE STUDIES OF FACTORS INFLUENCING THE GROWTH OF CHLORELLA PYRENOIDOSA

EDWIN A. DAVIS

During the year laboratory investigations have been carried out to study quantitatively the influence of various factors on the growth of *Chlorella*. It was the purpose of these investigations not only to find ways of increasing yield, but also to develop better culturing methods and to gain a clearer understanding of the effect of the factors studied on the growth process.

Night temperature and aeration. Night temperature is known to be an influential factor in the growth of many plants. Although the optimal temperature for *Chlorella pyrenoidosa* (Emerson's strain) when grown under continuous illumination is known to be approximately 25° C, the thermoperiodicity pattern required for op-

timal growth under day and night conditions was not known. Growth, measured by increase in fresh weight and cell number, was therefore studied as a function of night temperature. With a day temperature of 25° C and approximately 1000 f.c. illumination (about one-tenth sunlight), the night temperature was varied from 5° to 35° C.

Increase in fresh weight was not greatly influenced by night temperatures of 10°, 15°, 20°, or 25°. The greatest increase occurred with a 15° night, in which case the yield was 106.1 per cent of the yield of control cultures kept at 25° continuously. Growth was retarded with 5°, 30°, and 35° nights. With 5° nights it was 94.8 per cent of the controls, and with 30° and 35° nights it was 88.7 and 60.8 per cent respectively. The influence of night temperature on cell number was of the same general nature, although at the highest and lowest temperatures the suppression of growth was much more pronounced than at intermediate temperatures.

Aeration at night had little influence on the yield, but it increased cell division considerably.

Diurnal fluctuations in cell division and enlargement. During experimentation with the rocking tray it was noticed that over a period of several days cell division took place only at night, whereas cell enlargement was restricted to the day. The regularity of this pattern was striking for several days, but it did not continue. A study was made, therefore, to find whether the diurnal pattern of cell division and enlargement could be reproduced and the factors responsible determined.

Cultures were grown in the laboratory with 12 hours of light and 12 hours of darkness. During the day the cultures were grown at 25° C at 1000 f.c. and supplied with 5 per cent carbon dioxide in air. At night they were transferred to constant-

temperature chambers and aerated without 5 per cent carbon dioxide. Night temperatures were varied from 5° to 30° C.

Marked diurnal fluctuations in cell division and enlargement were found. During the day the cells showed an increase in size, followed by little or no further increase at night. This pattern was independent of night temperature. The time of cell division, however, was highly dependent on night temperature. For example, with night temperatures of 20°, 25°, and 30° C, cell division occurred almost entirely at night, but with 5° nights it occurred chiefly during the day. With the intermediate night temperatures of 10° and 15° it occurred during both the day and the night. The cultures which received 20°, 25°, or 30° C nights contained small cells in the morning as the result of cell division. Upon illumination, these small cells, now actively photosynthesizing, increased in size. By keeping the cultures under favorable day and night conditions, this process was repeatable.

The time of cell division could be restricted to the night hours by controlling the night temperature only so long as the cells received ample light during the day for photosynthesis. When the culture density increased to the point where many of the cells no longer received enough light, then division occurred during both the day and the night.

Culture medium. When Chlorella is grown under nonaseptic conditions or in alkaline media, the problem exists of maintaining an adequate supply of iron in a form available to the cells. Because of the difficulty of maintaining soluble iron in media containing ferrous sulfate or ferrous citrate when aseptic precautions are not observed, the commonly used chelating agent ethylenediamine tetraacetic acid (EDTA), which is biologically inert, was tested with a range of iron concentrations.

Very satisfactory results were obtained when iron was added as ferric potassium EDTA. Iron in this form was found not to be toxic in concentrations as high as 80 ppm, and is now being used routinely for nonaseptic culture work.

During the growth of Chlorella the pH of the medium changes greatly when either ammonium or nitrate ion is used as the nitrogen source. In order that such pH fluctuations might be avoided, a search was made for organic compounds which could be used as suitable nitrogen sources. Both glycine and urea were found to support excellent growth without causing drastic pH changes in the medium, urea being superior to potassium nitrate. Starting with a pH of 5.0, final pH values of the nitrate, urea, and glycine media, after 5 days, were 8.1, 6.5, and 4.9 respectively. When the urea medium was started at pH 6.0 it increased to only 6.4. Changes in the pH can therefore be kept within very narrow limits.

Neither urea nor glycine serves as a carbon source for *Chlorella*, a fact which permits these compounds to be used in investigations where carbon dioxide is to be the sole source of carbon. Urea, unlike glycine, does not support the growth of culture contaminants, and may therefore be used for nonaseptic as well as aseptic culture work.

From a study of the tolerance of *Chlorella* for potassium nitrate and urea it was found that twice as much nitrogen can be added as urea than as potassium nitrate without seriously decreasing growth. This fact will find application in the culturing of dense populations, where nitrogen rapidly becomes limiting and it is desirable to add as much as possible at a time without decreasing growth.

Urea therefore has several advantages over potassium nitrate as a nitrogen source for *Chlorella*: it gives more rapid growth;

it causes only minor pH fluctuations in the medium; and it can supply more nitrogen at one time without seriously decreasing growth.

Culture turbulence as a means of increasing yield. Sunlight is at least ten times as strong as the lowest light intensity that will produce maximum growth of Chlorella. In order to obtain substantially increased yields it is therefore important that more of the available sunlight be used for photosynthesis. From photosynthetic investigations it was shown that it should be possible to increase yields by subjecting individual cells to very short flashes of bright light followed by dark periods about ten times as long. For increased yields, however, it is necessary that when light does not fall on any given cell, it fall on another. A possible way of accomplishing this is by means of culture turbulence. An experiment was performed, therefore, to find whether increased yields could be obtained by this means.

A special apparatus was constructed which permitted the cells to be cultured in a thin layer (6.4 mm) exposed to bright light, approximately equivalent to sunlight, and subjected to various degrees of turbulence. Culture turbulence was accomplished by means of a spinning stainless steel rotor suspended in the culture, forming its inner boundary. By spinning the rotor at different speeds various degrees of turbulence were achieved. Carbon dioxide in air was introduced through the bottom of the chamber and bubbled up through the culture.

The influence of turbulence on growth was determined at the very high culture density of about 150 g/l fresh weight. The cells were transferred to fresh medium every 12 hours in order to provide ample nutrients and to remove any inhibitory excretory products that might be produced. The light was provided by six 300-watt re-

flector spot lamps equally spaced in a circle with their front surface 20 cm from the culture. Approximately 7000 f.c. was thus provided at the culture surface, and nearly all the light was absorbed in a one-millimeter layer of the suspension.

At the highest speeds, the culture was subjected to considerable turbulence. At o rpm it was stirred only by the gas bubbles. Growth, in terms of grams dry weight per square meter per 12 hours at the various rotor speeds, was: o rpm, 25.2; 16 rpm, 34.1; 208 rpm, 42.8; and 475 rpm, 43.2. It is seen that by increasing the rotor speed from 0 to 475 rpm a 71 per cent increase in growth was obtained. Growth at o rpm was about twice the 12 g/m² commonly found in experiments with other types of culture units, but the temperature control and the frequent provision of fresh medium probably account for this. It may be that an effect of flashing light greater than that noted can be found at other culture densities.

dioxide concentration Carbon growth. Since carbon dioxide is one of the most important factors that regulate photosynthesis and thus algal growth, it is essential that our knowledge of its influence be as complete as possible. In Chlorella culture, the answer to the question, What is the lowest prevailing carbon dioxide concentration in the culture solution which will support maximum growth? is of considerable importance. A study was made, therefore, to determine the influence of carbon dioxide concentration on the growth of Chlorella at several light intensities.

The culturing apparatus was built by Myers. It consists of a thin lucite culturing chamber fitted with an outside overflow tube which extends from the bottom of the chamber to the surface of the culture, and a photoelectric control system for maintaining the culture at a constant cell density by regulating the input of fresh me-

dium. As medium is added, some of the culture passes out of the overflow tube. The volume of overflow collected in a given time is a direct measure of the growth rate. A built-in water chamber provides temperature control. By means of this device it is possible to maintain constant culture density, pH, light intensity, temperature, and carbon dioxide concentration, and to set any of these factors at the desired value. Growth rate under any given set of conditions was usually determined over 24 hours. The carbon dioxide concentration with which the culture was in equilibrium was measured by the Van Slyke method.

The influence of carbon dioxide concentration on growth was studied at four light intensities: 120, 350, 950, and 2500 f.c. Growth increased with light intensity up to somewhere between 500 and 800 f.c. At all intensities maximum growth was achieved at carbon dioxide concentrations slightly greater than 0.03 per cent, growth with air alone being about 82 per cent of that with 5 per cent carbon dioxide in air. It is imperative, however, that when these low concentrations are used, the culture be kept saturated with the gas, which must therefore be bubbled through the culture very rapidly.

It appears that under conditions of light saturation and low carbon dioxide concentrations (i.e., below those necessary for CO₂ saturation), growth is limited by the rate at which the CO₂ acceptor becomes saturated; but below light saturation and CO₂ saturation, both the light reaction and the rate of saturation of the CO₂ acceptor limit growth.

PHOTOSYNTHESIS IN FLASHING LIGHT BESSEL KOK

Efficiency measurements. Algae have been shown to be able to convert up to 25

per cent of absorbed light energy into organic materials during growth in relatively weak light. However, in none of the experiments in which algae have been exposed to natural sunlight, which is of far greater intensity, has the yield of conversion surpassed 2 per cent. A serious limitation on the yield per unit amount of light absorbed in the outdoor growth of algae, even with an ample supply of carbon dioxide, is the high intensity of sunlight during a large part of the day.

Photosynthesis, the basic process of light transformation underlying algal growth, does not run faster and faster as more light is given, but finally reaches a certain speed, limited by internal cellular factors, typical for each strain of plant material and influenced by the prevailing temperature. Photosynthesis is known to be a chain process, a photochemical reaction being followed (or preceded) by several nonphotochemical steps, probably of enzymatic character. It is one of these so-called "dark reactions" which at a certain over-all rate will reach its maximum speed and in this way will impose a ceiling, the saturation rate, on the maximum rate of the whole chain.

We can consider the limiting step of this chain as a reaction in which definite numbers of enzyme molecules are at work transforming intermediate products from one form to another. At maximum speed, nearly all the enzyme molecules will be loaded and go through their cycle in a certain time. Only when such a molecule becomes free again can it be reloaded by a preceding, photochemically formed, intermediate.

When much more light is absorbed by the cells than can be handled in this reaction, only a fraction of the light will actually be used and its energy fixed in organic material. But if this same high intensity were given in short flashes, each flash short enough not to overshoot the amount of available enzyme molecules, and a dark period were interposed which was long enough to allow these molecules to discharge completely, no light would be wasted.

Of course, nothing would be gained in practice by periodically intercepting the sunlight by some mechanical device; the light losses, instead of occurring within the cells, would occur outside. This difficulty might be avoided by use of very dense cultures of algae stirred so that the cells would alternately shade one another, thereby exposing themselves to alternate light and dark periods. The population density and turbulence of the culture could be adjusted so as to provide the ratio of light and dark periods necessary to effect maximum photosynthesis by the individual cell.

The mere "spreading out" in time of bright light over more cells so that each gets on the average no more light than it can handle is, however, not sufficient of itself to ensure maximum photosynthetic efficiency. The timing schedule must also fit the cell's requirements. Only in case the light-dark cycles are shorter than a given time (determined by the limiting steps described above) can substantial increase in yields be expected. In order to furnish basic data regarding the intermittence pattern of illumination required for the engineering design of improved devices suitable for mass culture of algae, measurements have been made of the photosynthetic rates of thin algal suspensions in intermittent light, produced by a rotating disk, as compared with their rates in continuous light with intensities up to and greater than that of sunlight.

The cells reached their maximum capacity for photosynthesis when illuminated with light of average intensity 10 per cent of that of full sunlight (saturation in-

tensity). Comparable cells grown outdoors utilize about 2 per cent of solar radiation, which corresponds to about 20 per cent utilization of light of saturation intensity. This agrees well with the 25 per cent conversion obtained in growth experiments under the most favorable conditions.

For the algal strain most commonly used, both in this laboratory and for large-scale culture, more or less complete sets of intermittence data were obtained at different temperatures. Two examples of data obtained at 30° C are given here.

In order to obtain the highest possible efficiency of light utilization in photosynthesis, the rotating disk had to be turned at about 3000 rpm, with the openings of the disk set so that the ratio of light to dark periods was 1 to 10. The cells under these circumstances were illuminated 2 milliseconds and darkened 20 milliseconds. The maximum efficiency was halved when the rotation was decreased to 600 rpm, making the illumination time 10 milliseconds and the dark time 100 milliseconds. Even with this intermittence regime, the yield per unit of absorbed light was five times that obtained with continuous light of the same intensity as that of the flashes.

The general conclusion is that fundamentally the possibility exists of raising the efficiency of utilization of sunlight by subjecting algal cells to the proper regime of intermittence of illumination, but that the maximum realization of this objective in large-scale developments through increased turbulence of algal cultures would be difficult technically and unsound economically.

GROWTH OF CHLORELLA IN INTERMITTENT LIGHT

J. NEAL PHILLIPS, JR., AND JACK MYERS

It is considered possible that in the mass culture of algae under sunlight illumination, increased yields may be obtained by turbulence of the culture suspension, which would provide intermittent illumination of individual cells of the suspension. The efficiency of the photosynthetic process in the utilization of high light intensity is known to be increased if the light is presented in very short flashes separated by sufficiently long dark periods. Since growth of an alga results from its photosynthetic activity, it is expected that effects of intermittent illumination on growth will also be observed.

The specific growth rate of *Chlorella* pyrenoidosa has been measured in unilateral illumination provided by a 1000watt projection lamp through a focused optical system. The image of a condenser lens was focused on the algal growth chamber by a second lens. With the lamp operated at 105 volts, the system provided 25 × 10⁴ ergs/cm²/sec of visible light energy as determined by thermopile measurements with and without an RG8 filter. Various intensities of continuous light were obtained by the use of neutral screens or neutral Wratten filters. Intermittent light was obtained by chopping the light beam with a sector placed very close to the condenser lens and driven through a gear reducer by a synchronous motor. Frequency and length of the light and dark periods were varied by the use of different gear reducers and different sector patterns.

Rate of growth was measured by a constant-density, constant-volume method. The lucite growth chamber was fashioned as an absorption cell of 6 cm internal diameter and 1 cm thickness cemented between two water cells for temperature control at 25° C. Constant density of the suspension was maintained at about 0.6 mm³/ml by automatic dilution controlled by a photometric device. The constant volume was determined by the position of an overflow tube. The volume of overflow per day di-

vided by the culture volume in the chamber gives the specific daily growth rate in loge units.

In continuous illumination the growth rate increases linearly with light intensity at low intensities, attains a value of 1.9 at 2.0 × 10⁴ ergs/cm²/sec, and then increases only slowly to 2.1 at 25 × 104 ergs/cm2 sec. The intensity of tungsten light at approximate light saturation, 2.0 × 10⁴ ergs/cm²/ sec, corresponds to 500 f.c. as measured by a calibrated Photronic cell. Because of the higher color temperature of sunlight, however, the corresponding intensity in footcandles would be somewhat higher for sunlight illumination. The linear relation at low light intensities between growth rate and light intensity shows an anomaly in that it appears to extrapolate to a finite growth rate at zero light intensity. The anomaly will be investigated further.

In intermittent light the data may be treated by comparing the growth rate produced by short flashes of high intensity with the growth rate under an equivalent amount of energy supplied continuously at a lower intensity. The growth rate under these two conditions was the same when the light of high intensity was given in flashes of one millisecond. The reality of the expected effect of intermittent light on growth, similar to that on photosynthesis, has been demonstrated, since the high intensity given in flashes was used as efficiently as the low intensity given continuously, whereas the high intensity given continuously was used very inefficiently. With longer light flashes of 4 and 16 milliseconds, the growth rate per unit integrated intensity becomes progressively lower. Data are being obtained for still longer light flashes, to provide quantitative description of the character and limits of the phenomenon.

A High-Temperature Strain of Chlorella

CONSTANTINE SOROKIN AND JACK MYERS

Recent studies on the mass culture of algae have demonstrated difficulties in restricting the temperature rise in dense suspensions under sunlight illumination (Year Book No. 50, 1950–1951, pp. 130–131). Many algae, including the *Chlorella pyrenoidosa* used in mass-culture studies, appear to be low-temperature organisms with temperature optima at 25° C or less. An alga with a higher temperature optimum would have practical advantages. A search in this direction has led to the isolation of a number of strains of *Chlorella* with higher temperature optima.

Samples of warm surface waters obtained from the vicinity of Austin, Texas were used to inoculate test-tube cultures in a Knop's solution at pH 6.8. The cultures were aerated with 4 per cent carbon dioxide in air, illuminated at 500 f.c. by fluorescent lamps, and thermostated at 32° C. After several serial transfers, samples of the suspension were plated out on inorganic nutrient agar, and bacteriafree cultures were established from singlecolony isolates. Preliminary study of twelve cultures led to the selection of one of these (Tx 1105) that showed rapid and continued growth at 39° C. After repeated culture at 30° C, another plating-out yielded a second series of isolates. One of these (Tx 71105) was selected for further study. Subsequently a third series of isolates was obtained directly from accumulation cultures at 39° C. Among the 61 cultures established, there are small but possibly significant differences in growth rate and physical characteristics. Cells of the various cultures are not readily distinguishable from the Emerson strain of Chlorella pyrenoidosa and are tentatively

regarded as strains of a species of Chlorella.

Tx 71105 and *C. pyrenoidosa* have been submitted to a comparative study of the effect of temperature on the growth rate at light saturation. Growth in test-tube cultures in photothermostats was determined in terms of the optical density measured by an Evelyn colorimeter with a 600-millimicron filter. The logarithm of the optical density plotted against time yields a straight line, the slope of which is the specific growth rate. Cultures at each experimental temperature were continued by serial transfer until the growth rate remained constant over two successive attempts.

Light saturation for growth of *C. pyrenoidosa* could be achieved by two banks of two 20-watt daylight fluorescent lamps delivering about 500 f.c. to opposite sides of the test-tube cultures. For Tx 71105 this arrangement was adequate at temperatures below 26° C. At higher temperatures, two banks of eight closely spaced 60-watt tungsten lamps delivering 1600 f.c. to opposite sides of the cultures were used to obtain light saturation.

At light saturation for *C. pyrenoidosa*, the growth rate was 0.7 log₁₀ unit per day at 22° C, 0.9 at 25.5° C, and 0.4 at 28.5° C, indicating a temperature optimum at about 25° to 26° C. For Tx 71105 the growth rate was 1.0 at 25.5° C, 1.6 at 28.5° C, 2.6 at 35° C, 2.7 at 39° C, and 2.3 at 41° C, indicating a temperature optimum at 38° to 39° C. It will be noted that the new strain Tx 71105 not only has a higher optimum temperature, but at this temperature has a growth rate three times as high as that of *C. pyrenoidosa*.

Preliminary manometric studies on Tx 71105 at 39° C have yielded maximum values of about 5 mm³ O₂/mm³ cells/hr for respiration and 100 mm³ O₂/mm³ cells/hr for photosynthesis. This is the

highest rate of photosynthesis per unit quantity of cell material found in any organism so far observed.

GROWTH OF A HIGH-TEMPERATURE STRAIN OF CHLORELLA AT HIGH DENSITY

HAROLD W. MILNER

Strains of *Chlorella* that thrive at considerably higher temperatures and have higher growth rates than *Chlorella pyrenoidosa* have been isolated by Sorokin in Myers' laboratory at the University of Texas, as reported above. Time was not available to test one of these high-temperature strains under outdoor conditions of growth. But, to obtain some idea of how a high-temperature strain might perform in comparison with ordinary *Chlorella* under the high cell density conditions visualized for mass culture of algae, preliminary experiments were conducted with flask cultures.

Two liters of culture in a 3-liter Fernbach flask gave a culture depth of 7.5 cm. The cultures were illuminated from below with incandescent lamps and were supplied with 5 per cent carbon dioxide in air. Cultures of the high-temperature strain Tx-S-11-5 were maintained at $39^{\circ} \pm 1^{\circ}$ C, and C. pyrenoidosa cultures were kept at $25^{\circ} \pm 1^{\circ}$ C, both with continuous illumination. The culture medium was the same as that used in the outdoor experiments described above.

The logarithmic growth-rate constant for the high-temperature strain exceeded the constant for ordinary *Chlorella* only in the range of cell density where the cultures were virtually transparent. As soon as the density of the cultures became great enough to cause absorption of an appreciable fraction of the incident light, whether at 1300 or 4000 f.c. intensity, the two strains of *Chlorella* yielded the same number of grams of cells per unit area per day. That

is, complete light saturation of the cells appears necessary for the high-temperature strain to show a higher growth rate than ordinary *Chlorella*.

From these preliminary experiments it appears that the greater growth rate of the high-temperature *Chlorella* in very dilute cultures will be of no advantage in mass-culture applications, where cell densities at least high enough for complete light absorption will be employed.

For mass-culture operations the high-temperature strain has the tremendous advantage, however, that it should at high cell densities produce the same yield at 39° C as can be obtained at 25° C with the ordinary *Chlorella*. The requirement for cooling on a large scale is thereby greatly reduced.

Before the relative advantages of the two strains for mass culture can be stated more definitely, more extensive and quantitative tests are needed, particularly under the actual operating conditions to be employed.

Mass Culture of Algae for Exploratory Analysis

ROBERT W. KRAUSS

In support of the program concerned with large-scale culture of Chlorella it appeared desirable to culture other species of algae in order to determine their adaptability to techniques of mass culture and to produce samples large enough for exploratory analysis. Though the species of Chlorella have the most rapid growth rates known for algae at present, and are therefore most promising with regard to bulk production, it is possible that other algae with somewhat slower rates of growth may contain organic compounds peculiar to them which may be of particular economic importance. Therefore a program for growing and screening a number of species

was initiated in co-operation with the Department of Botany at the University of Maryland.

Cultures of 70 species and strains of algae on agar slants were obtained from the collections of Dr. E. W. Fager, Dr. E. G. Pringsheim, and Dr. C. B. van Niel. Pure cultures of species selected from this collection were grown in liter flasks under artificial light in the laboratory. These cultures served as the inocula for the mass-culture apparatus installed in the greenhouse.

Two types of culture vessel were employed: thirty 18-liter glass carboys, and three 400-liter Tygon-coated plywood vats. The carboys were closed with rubber stoppers equipped with filters containing gas inlets and outlets. The bubbling of a 5 per cent carbon dioxide—in—air mixture released at the bottom of the carboys served to prevent sedimentation. The vats were lined with 0.004-inch polyethylene sheeting and covered with sheets of plate glass. The culture in each vat was circulated by means of two motor-driven stainless steel propellers mounted in diagonally opposite corners.

A modified Knop's solution with iron added as citrate and supplemented with the micronutrients calcium, boron, zinc, copper, magnesium, molybdenum, and cobalt served as the nutrient solution. The pH was adjusted to conform to the known requirements of the species under culture, and was maintained near the optimum by the periodic addition of nitric acid. The total delivery of the 5 per cent carbon dioxide-in-air mixture supplied to the cultures was 1000 liters per hour.

During the intense summer heat in the greenhouse the carboys were cooled by water seeping over them from a porous canvas hose laid across the shoulders. The water then flowed along the trough in which the carboys were standing, effecting

additional cooling. This system made it possible to hold the temperature of the cultures as much as 20° C below the prevailing air temperature. The vats presented a different problem. Reduction of the temperature could be obtained by removing the plate glass covers; rapid evaporation then kept the cultures below 40° C on all but the hottest days, so that no cultures were lost from overheating. Reduction of growth at these temperatures, however, coupled with the impossibility of maintaining a saturated 5 per cent carbon dioxide-in-air atmosphere over the surface of the uncovered vats, made this arrangement unsatisfactory. Inasmuch as no efficient heat-conducting surface came in contact with the culture solution, two stainless steel tubes 3/4 inch in diameter and 5 feet in length were placed in each vat. Cold water from the mains flowing through the tubing served to hold the temperature as much as 15° C below that in the vats not so equipped. The hotter the day, the greater the temperature differential observed.

Inasmuch as the algae were grown in a nonsterile system from the time of inoculation from the laboratory cultures, considerable care was taken to ensure the maintenance of unialgal cultures. A minimum of 100 ml of pure inoculum containing at least 40×106 cells/ml was introduced into each carboy; each vat in turn was inoculated with the contents of one carboy after it reached a similar concentration. The heavy initial inoculation minimized the possibility that another species might supplant the desired one in the medium. The carboys and all apparatus coming in contact with the cultures were either autoclaved or soaked in 5 per cent formaldehyde solution between cultures. The 0.004-inch polyethylene sheeting lining the vats was replaced with fresh sheeting whenever a new species was introduced. Species with distinct morphological differences were cultured in adjoining vats whenever possible, so that cross-contamination could be more easily detected. No cross-contamination or infestation by "wild" species of algae was detected by daily microscopic checks. Bacteria were present, but did not appear to injure the cultures. Several vats were infested by a ciliate too small to ingest the algae. Though no corrective measures were applied, the ciliates disappeared after several days.

Growth was measured by centrifuging at 2000 rpm (RCF-1160) for 20 minutes in 100-ml Goetz centrifuge tubes graduated in hundredths of a milliliter, and by cell count using a hemacytometer. The species differed in cell size, so that no ratio of packed cell volume to cell count could be applied to all. Packed cell volume (pcv) was accepted as the primary index of growth. Inasmuch as light and temperature were variables, no absolute maximum growth rates could be determined. The weight reported in column three of table 5 is the average daily fresh-weight yield per liter of culture during the entire growing period from inoculation to harvest. It includes the early lag phase as well as the period beyond that of logarithmic growth. The yields reported for the carboys are invariably higher than those reported for the vats. The vats, however, have only 50 cm² of illuminated surface for each liter of culture, as contrasted with 220 cm² per liter in the carboys. The vats compare more favorably when illuminated surface per unit volume is used as a criterion.

Harvesting was accomplished by pumping the culture through a Sharples Super Centrifuge. Carboys were harvested when they reached a concentration of 0.8 ml/100 ml pcv. The supernatant medium was discarded and fresh medium was added to each carboy, in which one liter of the

culture was retained as an inoculum. The vats were harvested when they reached a concentration of 0.2 ml/100 ml pcv. The algae were removed until the culture was reduced to a concentration of 0.1 ml/100 ml pcv. Additional salts were then added to compensate for those used by the algae; this permitted a continuous system of culture.

Thus far, attention has been principally devoted to the species and strains of Scenedesmus. It is known that S. obliquus, strain D₃ of Fager, contains chondrillasterol, a sterol having a structure which makes it promising for the synthesis of cortisone. It appeared desirable to examine as many species and strains of Scenedesmus as possible, to determine which contain valuable sterols and which produce the highest sterol yield. Dr. W. J. McAleer, of Merck and Company, Inc., is analyzing the samples produced for their sterol content. To date, the most promising organisms are two strains of S. obliquus, D3 of Fager and WH-50 used in previous work at the University of Maryland. The sterol yield for D₃ was 0.2 per cent of the dry weight, as compared with o.i per cent of the dry weight for WH-50. The sterols in the two strains are not the same, but the molecular structure in both suggests a likely route to cortisone.

In an effort to determine the effect of a reduced inorganic nitrogen supply on the sterol content of the algae, a harvest of *Scenedesmus obliquus* D₃ was transferred to a medium without nitrogen and maintained there for 14 days. The sterol content of these nitrogen-deficient cells proved to be no greater than that of the algae grown in the nitrogen-rich medium.

Additional analytical work to determine the amount and nature of the organic phosphates found in these algae has been undertaken by Dr. F. P. Veitch, of the Chemistry Department at the University of Maryland. The culture of the algae has been carried on with the technical assistance of Mr. Robert M. Latane and Mr. Donald H. Boughton.

measurements have been made of the freesettling rates of *Chlorella pyrenoidosa* under different conditions.

When a homogeneous suspension of

TABLE 5

Total harvest weight and yield of Scenedesmus species grown in mass culture

Species and source	Total fresh wt. (g)	Yield, fresh wt (g/l/day)	
S. basiliensis (Pringsheim 276/1):			
Carboys	857	0.281	
Vats	1,187	0.069	
S. basiliensis (Pringsheim 276/16):			
Carboys	1,201	0.232	
Vats	968	0.073	
S. bijugatis (Van Niel Z.14.2.1):			
Carboys	821	0.285	
Vats	1,015	0.012	
S. Naegelii (Pringsheim 276/2):	,		
Carboys	1,216	0.239	
Vats	1,272	0.059	
S. obliquus (Fager D ₃):	,		
Carboys	2,647	0.150	
Vats:	,		
Nitrogen +	1,696	0.080	
Nitrogen	1,028		
S. quadricauda (Pringsheim 276/4b):	-,		
Carboys	821	0.244	
Vats	994	0.059	
S. sp.* (Van Niel Z.14.3.1):			
Carboys	834	0.224	
Vats	707	0.084	
S. sp.* (Van Niel Z.14.4.1):			
Carboys	816	0.272	
Vats	550	0.115	
Total	18,630		

^{*} Not identified.

Sedimentation Rates of Chlorella Pyrenoidosa

JAMES H. C. SMITH

Continuous harvesting by sedimentation has been proposed as one means for concentrating the cells of *Chlorella* in the large-scale production of this organism. In order to supply numerical data for estimating the performance of such a scheme,

Chlorella cells, contained in a cylinder, was allowed to settle, the free-settling cells produced a well defined boundary. The downward displacement of this boundary with time served as a measure of the settling rate of the cells.

The measurements showed that the displacement of the boundary was directly proportional to the time. The rate of displacement of the boundary was a function of the cell content of the suspension and obeyed approximately an equation of the form $R = K/C^n$, where R is the rate of displacement of the cell boundary in centimeters per hour, C is the initial cell content of the homogeneous suspension in milligrams per milliliter, and K and n are constants. The greater the cell content, the slower the downward movement of the boundary and the greater the quantity settled in a given time. Individual cells differ in their rate of settling. Cells which had been previously sedimented by a light centrifugation settled faster than those which had remained suspended under the same conditions.

In one experiment, the constants of the equation were K=0.143 and n=0.292. On a fresh-weight basis, it is estimated from the equation that a kilogram per day will settle through a distance of 1.75 cm over an area of 5.7 m² when the cell content of the suspension is 10 g/l.

Multiple Culture Apparatus for Algae Robert W. Krauss

During the progress of experiments involving algae in mass culture, the need for a supplementary apparatus designed to handle many smaller cultures became apparent. Questions concerning optimum temperature, toxicity levels, comparative effectiveness of nutrient solution, carbon sources, and the like could not be readily answered unless cultures were adequately replicated and maintained under constant conditions. A continuous culture system aimed at a steady state during the entire course of growth is ideal. Nevertheless, introductory information can be more rapidly obtained by holding all environmental conditions constant except those modified by the growth of the algae themselves.

The device developed for this purpose

is essentially a shaker for rocking culture flasks in a thermostatically controlled water bath. Ten 500-ml Erlenmeyer flasks immersed to three-quarters of their depth in an aquarium-type water bath are rocked 90 times per minute through an arc of 20°. The rocking motion for each of two sets of five flasks is transmitted to the clamps holding the flasks by a pitman attached to a cam. The pin bearing the pitman on each cam may be adjusted to permit variation in the rocking motion. By setting the pin near the center of the cam, rocking through an arc as small as 2° can be obtained; setting the pin near the circumference of the cam increases the arc while maintaining the same number of revolutions per minute. The velocity of revolution may be varied by exchanging the sprockets on the 1/12-horsepower gear-reduction motor which provides the power. The cultures may therefore be rocked gently with little or no turbulence, or agitated vigorously to produce a strong churning action.

The water bath surrounding the flasks is 75 cm long, 50 cm wide, and 24 cm deep. The flasks are suspended with their bases 10 cm above the glass bottom of the tank. Temperature is controlled by a thermostat and an electric heater which warms the water flowing from one end of the tank to an overflow tube at the other. Wave action in the bath is minimized by setting the pin on one cam 180° from that on the other. This allows one set of flasks to move in a direction opposite to that of the other.

Illumination is provided by batteries of incandescent and fluorescent lights mounted on a movable steel frame below the glass bottom of the water bath. The intensity and wave length of light can be varied by changing the lamps or by changing the level of the steel frame. The entire mechanism, mounted on an angle-

iron frame, may be covered to exclude extraneous light.

Any desired mixture of carbon dioxide in air may be metered by Fischer-Porter flowmeters calibrated in liters per hour. The gas is delivered to a manifold equipped with a needle valve for each flask. Each valve is connected by rubber tubing to a glass tube extending through a rubber stopper to just above the bottom of the flask. Both inlet and outlet are equipped with sterile cotton and glass wool filters so that pure cultures may be maintained.

The apparatus was constructed with the co-operation of Professor George J. Burkhardt, of the Department of Agricultural Engineering at the University of Maryland.

Oxygen Production by Chlorella in the Absence of Atmospheric Carbon Dioxide

EDWIN A. DAVIS

Illuminated Chlorella cells suspended in glucose-Knop's solution and deprived of atmospheric carbon dioxide liberate oxygen. Most of the oxygen liberation is indirectly caused by nitrate reduction, for which glucose and phosphate are required (see Year Book No. 49, 1949-1950). If, however, the cells are suspended in distilled water containing glucose, thereby eliminating the possibility of nitrate reduction, oxygen production is still observed although at a slower rate than when nitrate is being reduced. In the absence of glucose, oxygen liberation is extremely slow and eventually stops. The ability of Chlorella, deprived of atmospheric carbon dioxide but supplied with glucose, to liberate more oxygen than the amount necessary to compensate for respiration is of particular interest. It indicates that there is a mechanism for the utilization of the hydrogen derived from water, by the photochemical step in photosynthesis, which does not rely upon the uptake of carbon dioxide. Possible mechanisms for the utilization of this hydrogen are the conversion of acetate into fats, and the reduction of intermediate oxidation products.

The concept that hydrogen derived from photochemical reactions can be used in cellular processes not involving the uptake of carbon dioxide is one which is worthy of considerable attention.

Reversion of a Photosynthetic Chlorella Mutant

EDWIN A. DAVIS

By irradiating wild-type Chlorella cells with ultraviolet light, mutants were produced which are unable to photosynthesize yet contain what appears to be normal chlorophyll. They grow, however, if supplied with a source of reduced carbon (see Year Books Nos. 49, 1949-1950, and 50, 1950-1951). The mutants are green when grown in either light or darkness, but in light after their supply of reduced carbon is exhausted they become bleached. Wildtype cells remain green under similar circumstances. The reappearance of green cells in bleached cultures is one method of detecting reversions to the wild type. Reversions have not yet been detected by this method for the two mutant strains unable to evolve oxygen, but have been found in the strain capable of producing oxygen.

Test tubes containing 0.2 per cent glucose in modified Knop's solution were sterilized and inoculated with the oxygen-producing mutant. The cultures were grown under fluorescent light. After growth ceased, the cells commenced to bleach. The bleached cultures were left exposed to light and examined periodically to see if the normal green color of any of the cultures was restored. Six months from

the date of inoculation, three out of four cultures were again green.

Transfers were made from the green cultures to minimal medium (modified Knop's solution) and to modified Knop's solution with 0.2 per cent glucose, care being taken to observe aseptic precautions. The cultures were then illuminated. Growth occurred not only in the glucose medium, but also in the minimal medium. None of the cultures bleached upon prolonged exposure to light. These facts indicate that the green cells which developed from the bleached mutant cells were no longer dependent on a supply of reduced carbon. They were capable of photosynthesizing.

This sequence of events has been repeated, so that there seems little doubt that the mutant is able to revert to the wild type. In the absence of quantitative data it is not known whether the reverts arose as the result of a mass reversion or of a single-cell reversion. Casual observation of the quantity of bleached cells before and after the restoration of the green color suggests that a mass reversion may have occurred. Reversions have never been observed during the ordinary tests of the ability of the mutant to grow in minimal medium, carried out for periods of only a few days. This may be because sufficient time has not been allowed for reversions, which were observed only in cultures kept for months after the exhaustion of the carbon supply.

Efficiency of Photosynthesis Bessel Kok

The conversion of light into chemical energy by photosynthesizing cells occurs discontinuously; each discrete energy step represents a transformation of a light quantum. For green cells, we can attribute an energy content of about 40 kcal per mole

to the energy level of excitation from which all chlorophyll-absorbed light quanta ultimately act.

The question of how efficiently green cells can bring about this conversion has both theoretical and practical importance. The theorist will prefer to express this optimal efficiency as the minimum number of light quanta necessary to evolve one molecule of oxygen or to reduce one molecule of carbon dioxide. If this number were low, it might lead toward a clearer understanding of the process.

The practical importance of knowing the optimal photosynthetic efficiency is that it would provide a basic figure for the conversion of solar energy into agricultural crops, mass cultures of algae, and the like. For this purpose, we prefer to express this yield as the ratio of chemical energy actually harvested in the form of organic material to the light energy absorbed during the period of growth. This ratio was determined earlier by us with Chlorella, by use of oxygen evolution during growth and chemical analysis and heat of combustion after harvesting as measurements of the amounts of energy fixed. Under optimal conditions 25 to 30 per cent yields were obtained, corresponding to quantum numbers of about 8 hv/O2 during the growth period. This figure, obtained from growth measurements, has met with more or less general acceptance.

The same cannot be said about the "theoretical" aspects. In the late thirties several investigators questioned the correctness of Warburg's early (1922) finding of a quantum number of about 4 hv/O₂, and a controversy has developed on this point. On one side, Warburg and collaborators have extended their experiments and have claimed to obtain optimal values of 3 quanta per O₂ evolved. On the other side, several groups, and notably Emerson,

have defended a value of 8 to 10 as the best obtainable.

A great part of this controversy concerned difficulties of measurement inherent in the use of "two vessel" manometric methods. More recently, however, Warburg has showed that his results could be equally well obtained in "single vessel" experiments. Therefore, the applicability of the two-vessel method will not be discussed here.

In earlier work, Kok showed that both 4 and 8 quanta could be found, depending upon the conditions of the experiment. When thin, partly absorbing *Chlorella* suspensions were irradiated so that all the cells were evenly and constantly illuminated, it was frequently found that the curve relating incident intensity to photosynthetic rate was composed of two straight lines of different slope, with the break at the so-called compensation point (i.e., the light intensity at which oxygen exchange is zero). The two slopes indicate two different quantum yields.

From this finding it was concluded that an interaction between photosynthetic and respiratory processes existed, and that whereas 4 quanta (under conditions as yet not fully understood) may be sufficient to suppress uptake of O2 in respiration, 8 quanta are needed for actually setting free an O2 molecule in "photosynthesis." Warburg and Burk rejected this proposition on the basis of experiments in which two light sources were used, the first being of unmeasured intensity and giving a diffuse pattern of irradiation which served to raise the whole level of O2 exchange of the suspension under measurement up to or over the compensation point. The other light source as usual was a narrow collimated beam of measured intensity. Again quantum numbers of 3 to 5 could be computed from the increased over-all O2 evolution brought about by the addition of the measured light.

The closer study of this point of controversy was the main objective of Kok's visit to Burk's laboratory, National Cancer Institute, National Institutes of Health, Bethesda, Maryland. Several other points also were examined during a month of co-operative work, some of which will be briefly discussed here.

The quantum numbers obtained were scattered over a wide range, from 3 up to high values. The question is, how much of this scattering is attributable to physical methods and how much to variation of the actual efficiency of the cells?

Although the manometric methods, and even more the optical methods, were not highly precise, and although the cursory examination of many points did not permit the attainment of very accurate absolute values, such values were not necessary for distinguishing between the controversial quantum-yield values, which differed by a factor of two. The relative values obtained—that is, the manometric data from one sample of cells illuminated with different light intensities of accurately known relative values—were sufficiently reliable for settling the main point at issue. Therefore, the great scattering of the data must be attributed to variations within the biological material itself.

The experimental arrangement devised by Warburg and Burk, in which the dense algal cultures are illuminated by two light sources of different intensities and geometrical patterns and which has been employed in most of Warburg and Burk's recent work and many of the present experiments performed at Bethesda, furnishes complicated results. In this set-up, when the suspensions are shaken, the cells are moved between regions of very different light intensities, a circumstance which leads to intermittence effects impossible to evaluate.

It is not within the scope of this report to give a detailed analysis of the data obtained at Bethesda, but it can be stated that earlier observations both of Warburg and Burk and of Kok were reproduced and to a certain extent harmonized.

Some of the results may, however, be reported. No significant differences in efficiency were found between cells suspended in growth medium, pH 4.35, and those suspended in carbonate buffer mixtures 0.2 M, pH 8.8 (or 0.1 M, pH 9.2).

In all cases when a single light source was used, "low" quantum numbers only (3 to 5) were found at light intensities below the compensation point. Higher intensities, 2.5 µl quanta/cm²/min * and over, caused a severe drop in the yields, even though, in thick suspensions, only one-third to one-half of the respiration was compensated by these intensities. As had been observed earlier by Kok, only in thin algal suspensions at low intensity were "low" quantum numbers observed. Above compensation of respiration, they increased.

In the apparatus used at Bethesda, the changes in rate of photosynthesis produced by the changes in intensity of the small collimated beam were almost exactly equal whether or not auxiliary illumination was supplied by the overhead fluorescent light. The absolute values as well as the relation between rates and intensities of the collimated beam were identical. From our results, it is obvious that the response of green cells to light is not simple, and that data obtained by such an arrangement as the one described do not give a constant quantum yield which is independent of experimental conditions.

Though the results observed may be interpreted in various ways, as has been pointed out by Burk, Kok believes that

* A µl quanta represents as many light quanta as there are molecules in a µl of gas under standard conditions.

they can best be explained by a reduction in respiration induced by the light. The interaction of green-cell respiration and light is in Kok's opinion the cause of the differences in values obtained for the quantum yield.

This interaction between light and green-cell respiration, though not fully understood as yet, either biochemically or in relation to its governing factors, may be the main stumbling block in the quantum-yield controversy.

Finally, it may be worth while to report on observations made in Bethesda concerning the response time of the manometric equipment used. After the intensity of illumination of the algal suspension in a manometric vessel was changed, several minutes elapsed before the new steadystate rate was reached. Roughly, it took from 1 to 5 minutes for half the transition between rates to occur. These observations do not significantly affect measurements on over-all photosynthetic yields, but they do bear directly on measurements considered to uphold the "one quantum" theory of photosynthesis proposed by Burk and Warburg, in which intermittence periods and reading intervals of I minute were employed. The slowness of response of the apparatus may make necessary a reinterpretation of the experiments leading to the proposed "one quantum" mechanism of photosynthesis.

PRODUCTION YIELDS OF GRASS JAMES H. C. SMITH

In last year's report, preliminary results were given on the production yields of the grass *Poa pratensis*, originally obtained from two localities, Groveland, California and Newport, Oregon. These experiments have now been extended so that data for a complete year's growth have been obtained. The two lots of grass in garden

soil were grown in a greenhouse in "flats" of 0.0632 m² area. They were given nutrient solution 5 or 6 days a week and usually harvested at from two- to fourweek intervals, depending on the rate of growth.

The yield in dry weight for the year, 366 days, was 161.2 g for the Groveland sample and 185.3 g for the Newport sample. These yields correspond to 2.55 and 2.93 kg/m²/year. The dry weights were 20.8 and 20.1 per cent of the fresh weights, respectively.

The rates of production were highest during April and May, the maximum rates being 12.7 and 13.3 g/m²/day. The lowest rates were in December, 2.26 and 2.94 g/m²/day. The average daily yields for the year were 6.97 and 8.01 g/m²/day.

FACTORS AFFECTING THE TRANSFORMATION OF PROTOCHLOROPHYLL TO CHLOROPHYLL

JAMES H. C. SMITH

During the past five years, experiments in our laboratory have demonstrated that chlorophyll is formed in the plant by photochemical transformation of protochlorophyll. This conversion takes place only within the plant: it has not been possible to accomplish the transformation with protochlorophyll extracted from the plant. In what combination the protochlorophyll exists inside the plant so as to make it readily convertible to chlorophyll, and by what kind of action it is converted whether by intramolecular rearrangement of the holochrome * or by intermolecular reaction mediated by enzymes—is still unknown.

* Holochrome (Gr. holos whole + chroma color) is proposed as a term to designate a colored substance as it exists in its natural state within an organism, where the colored prosthetic group is combined or associated with a carrier which alters the physical or physiological properties of the prosthetic group.

In the older literature, fragmentary knowledge is recorded which bears on these points, but much of this knowledge has been obtained under such experimental conditions as to make it either of questionable value or impossible of interpretation. Because of this, a series of experiments has been undertaken to establish the effects of various factors on this conversion. It is hoped that these experiments will make it possible to determine the nature of the reactants and of the reaction itself.

Temperature. Previously (Year Book No. 48, 1948–1949, p. 91) it was reported that the rates of conversion of protochlorophyll to chlorophyll at 5° and 18° C are identical and that the conversion is stopped by a temperature of 90° C (Jour. Amer. Chem. Soc., vol. 70, p. 3558, 1948). Further experiments have now been performed to determine the temperature limits of the reaction.

An estimate of the upper temperature limit was obtained in the following way. Etiolated barley leaves were immersed in distilled water at different temperatures for 5 minutes. After the leaves were removed from the water, they were illuminated for 15 minutes with light of 100 f.c. intensity. The leaves were immediately extracted with acetone and the extent of the conversion was determined spectrophotometrically (Arch. Biochem. and Biophys., vol. 31, p. 5, 1951). Treatment at 68.5° C stopped the conversion. At 51° to 52° C there was 35 per cent conversion, and at 48° to 49° C, 68 per cent conversion. Interpolation of these data gives a value of 50 per cent conversion after a 5-minute treatment at 50° C. This temperature is in the range of temperatures frequently found to cause inactivation of enzymes and denaturation of proteins. This suggests that the inhibition of this conversion might be ascribed to either or both of these causes.

In an attempt to determine the low-

temperature limit of this transformation, etiolated leaves of barley were frozen and illuminated at -43° and -76° C. At these temperatures the conversion was 72 and 35 per cent complete, respectively. The fact that the conversion occurs rapidly at such low temperatures in the frozen leaf is evidence against an enzyme-mediated reaction and evidence for a photochemically induced intramolecular rearrangement within the holochrome.

Although irradiation of the frozen leaf produced the conversion, irradiation of the thawed leaf produced no appreciable conversion. This lack of conversion might have been attributed to a selective loss of chlorophyll (had it been formed) by illumination of the thawed leaf in air, since the total pigment content of the leaf decreased greatly under these circumstances. It was demonstrated, however, that thawing and illuminating the leaf in an inert atmosphere, carbon dioxide or hydrogen, preserved the pigment almost quantitatively but inhibited the conversion completely. Since the conversion is known to take place under anaerobic conditions, it is concluded that thawing disrupts the holochrome in such a way as to make it photochemically inactive.

Drying. The controversial question whether this transformation takes place when dried leaves are illuminated has been re-examined. When etiolated barley leaves were dried in vacuo in the dark by means of calcium chloride or phosphorus pentoxide, or by freeze-drying (lyophilizing), they retained all or a large part of their protochlorophyll. Leaves that were thoroughly dried by means of calcium chloride or phosphorus pentoxide at room temperature transformed little or none of their protochlorophyll to chlorophyll when they were illuminated. When, however, the leaves were frozen and dried by lyophilization and then illuminated, they converted an appreciable fraction of the protochlorophyll to chlorophyll—in two instances, 46 and 28 per cent. The leaves had lost 90 per cent of their fresh weight during the freeze-drying.

The fact that the transformation takes place in the lyophilized state is more consistent with the assumption that the conversion is a photochemical rearrangement within the holochrome than that it is an enzymic process.

Infiltration. The photochemical conversion of protochlorophyll to chlorophyll in etiolated barley leaves is not greatly inhibited by infiltration with a number of substances, as is shown by the summary of results given in table 6. In each experiment the intensity of illumination was approximately 100 f.c., the time of illumination was about 15 minutes, and the retention of the pigment was very good.

Potassium cyanide, which complexes heavy metals and frequently inhibits respiration in living organisms, has little effect on the transformation when allowed to act for a short time. The same holds true for the protein precipitants ammonium sulfate and potassium ferricyanide, and the protein denaturants urethane and urea. Also the polyhydric alcohols are ineffective as inhibitors.

When, however, the protein denaturant urea acts for longer periods of time, its inhibitory action increases. The inhibitory action of urea cannot be reversed by soaking the infiltrated leaves in distilled water. The inhibition of the transformation by a protein denaturant implies that the active system involves a native protein.

Disintegrated leaves. The most significant results would be obtained if the system which undergoes this photochemical transformation could be isolated in active form from the plant and its nature determined. Such results would be of significance not only for understanding this

specific reaction, but also for understanding the photosynthetic mechanism, which is so imperfectly known. As one step toward the accomplishment of this objective, the ability of several substances to preserve an active system during grinding and subsequent illumination of etiolated leaves has been tested. Of the substances tested, glycerine is the most effective. Leaves

has been made in extending such experiments to the transfer of energy between extracted plant pigments. Unambiguous spectroscopic evidence has been obtained confirming the energy transfer from chlorophyll *b* to chlorophyll *a* in concentrated ether solution, an effect which was previously reported from other laboratories. It is likely that the light energy absorbed by

TABLE 6

Effect of infiltration of etiolated barley leaves with various substances on the conversion of protochlorophyll to chlorophyll a

Substance	Conc. (%)	Infiltration time (min)	Conversion (%)
Potassium* cyanide	0.065	90	89
Potassium ferricyanide	3.3	33	90
Ammonium sulfate	33	60	77
Glycerine	100	140	76
Sucrose	33	238	84
Sorbitol borate	80	60	74
Urethane	10	65	78
Urea	33	2	90
	33	65	63
	33	93	27
	33	240	15

^{*} Cf. C. S. Yocum, Amer. Jour. Bot., vol. 33, p. 828 (1946).

which were ground and irradiated in glycerine maintained their pigment content almost quantitatively and transformed approximately 70 per cent of their protochlorophyll to chlorophyll. The outlook seems promising, therefore, for the isolation of the active system from disintegrated leaves.

Fluorescence Spectra of Chloroplast Pigments

VIOLET K. YOUNG

The transfer of light energy from one chloroplast pigment to another in a living unicellular red alga as determined by fluorescence spectroscopy was discussed in last year's report. This year a beginning

chlorophyll b in ordinary green leaves goes through this transfer to chlorophyll a before being used in photosynthesis.

The fluorescence spectra of *Chlorella* suspensions of various cell densities and layer thicknesses have been measured. These measurements were made to obtain the action spectrum for the excitation of chlorophyll *a* fluorescence with sufficient precision to evaluate the influence of the other pigments present. Because of the reabsorption of the fluorescent light within the alga, the initial objective has not yet been achieved, but the use of cells grown so as to contain small quantities of chlorophyll are expected to make such measurements possible.

The distortions of the recorded fluores-

cence spectra due to selective reabsorption of the fluorescent light within solutions of chlorophyll *a* have been measured under various conditions. Corrections based on separately determined absorption spectra were applied to the distorted fluorescence spectra, permitting successful calculation of the true fluorescence spectra in the case of solutions. This may show the way to applying similar corrections to the more complex situation in the fluorescence spectra of intact algal cells.

THE CURVE ANALYZER

C. Stacy French, Donald R. Bellis, and Richard M. Cook

The machine previously mentioned in the Year Book reports which was built for carrying out simple mathematical operations on curves has been improved and somewhat expanded. In spite of long interruptions for remodeling, it has been used for many different problems in the past year. Owing to the incorporation of a servo-driven integrator and provision of more flexibility in multiplying and adding, the device has become more of a generalpurpose computer than was originally visualized. It can make any equation, within its capacity, come alive by plotting the dependent variable against the independent variable when the latter is automatically swept through the desired range. This greatly facilitates the fitting of equations to data. Such use as a rudimentary analogue computer, as, for instance, in applying some equations of chemical kinetics to laboratory data, may perhaps be of as much value as its already extensive use has been in converting scales of curves and analyzing complex empirical curves by fitting them with algebraic combinations of simpler ones, as in the analysis of spectra of mixtures of pigments. Curves of common functions of x such as 1/x, e^x , e^{-x} , x^2 , x^3 , x^4 , $\log x$, $\log 1/(1-x)$, $e^{-0.5x^2}$ are kept on hand to be put into the machine and combined in various ways to plot simple equations. The constants and parameters are introduced by dial settings which are adjusted by trial until the resulting plot of the equation fits the experimental data, a graph of which has previously been put on the recorder table. The constants of differential equations are equally well evaluated in this way by making the machine operate so that the curve which it plots fits the data given graphically in integral form. Thus integral or differential equations of different forms or with different constants can be compared as to their fit to series of experimental observations without carrying through calculations for each individual observation. Some of Kok's data for photosynthesis rates in flashing light are thus being analyzed. The results have so far been of more value in showing up the potentialities and limitations of the curve analyzer than in advancing the kinetics of photosynthesis.

The necessary development from a device that worked nearly well enough to a reliable machine for general use has been difficult. Through the help of Mr. William R. Fair, on a consulting basis, most of the serious troubles have been eliminated. The stability and dynamic response of the tables, followers, and pen have been improved in the past year, and it now seems likely that the present arrangements will not have to be changed radically in the foreseeable future. The mechanical work on the last two of the five follower units has been completed. Owing to design changes, construction of the electrical components for the last two followers has been delayed and will require a few more months. New photocell carriages and the improved mechanical components of their

drive systems have been put on all the follower units. The follower system finally installed on all units uses small flashlight bulbs on the moving carriage and crosscoupled D.C. amplifiers that reduce the drift to a usable, although still undesirable, amount while providing satisfactory precision and response speed. The curves which the machine plots are now smooth, rather than ragged as they were before the thorough redesign and rebuilding of the follower, table, and computing amplifiers. It has therefore become possible to use a large, flat pen on the recorder to plot curves that can immediately be put into a follower unit without hand inking, although an inaccuracy of about 1/16 inch,

due to the variability of the ink meniscus, may be introduced.

The construction of a simple servo voltage multiplier with two coupled potentiometers operated by a system similar to the table drives is being planned. Operating instructions have been written and forms prepared to facilitate the machine set-up for different purposes. The remaining electrical construction must still be completed, adequate detailed performance tests made, and a description prepared for publication. Possible further expansion by addition of two more integrators may be considered later if further experience with the machine shows their need in the type of problem with which we are concerned.

PALEOBOTANY

RALPH W. CHANEY

During the past four years much attention has been paid to the history of Metasequoia and to its botanical relationships. Most of the fossil material available for our study was collected during the past century in western North America; it has been found that a majority of the specimens identified as Sequoia, and many of those considered to represent Taxodium, are properly referable not to these genera now living in the United States, but to the related Metasequoia of central China. (See Year Book No. 49, 1949–1950, pp. 114–116.) For many years coniferous foliage specimens from Greenland, Spitzbergen, and Grinnell Land have also been assigned to Sequoia and Taxodium, but the illustrations in the works of Heer and others have suggested that their relationship is with Metasequoia. Since the climatic and other physical requirements of these three genera are significantly different, and since broad conclusions regarding earth history have been based on the earlier identifications, it has seemed desirable to check as many of them as possible. In the early summer of 1951 opportunity was afforded to study the original collections from these northern islands in museums at Stockholm, Copenhagen, and London. All the specimens of Sequoia with distichous (flat-sprayed) foliage were found to be referable to Metasequoia, as were many of those formerly identified as Taxodium.

The dominance of Metasequoia at high northern latitudes during the Cretaceous and Tertiary periods seems fully established. Its widest distribution and greatest abundance were during the Eocene epoch, in early Tertiary time; thereafter it was restricted to middle latitudes, becoming extinct in North America at the end of the Miocene epoch, some fifteen million years ago, and lingering on in Japan into the Pliocene epoch, perhaps five million years ago. Approximately one thousand trees have survived in central China, in remote valleys not yet cleared of their forests to provide land for cultivation. This changing pattern of distribution in time

and space has been variously interpreted by students of earth history, and always on the mistaken assumption that an evergreen conifer, Sequoia, was involved. Earlier reports (Year Book No. 48, 1948-1949, pp. 106-107; No. 49, 1949-1950, p. 115) have mentioned the significance of the deciduous habit of Metasequoia: most trees which shed their leaves annually are found in regions of summer rainfall and low winter temperature. Planting experiments with seedlings are showing that Metasequoia can survive the winters of southeastern Alaska, and confirm evidence from other sources that Cretaceous and Tertiary climates at high northern latitudes were temperate rather than tropical as has been so often suggested.

If the shift in distribution from high to middle latitudes represents forest migration resulting from changes in climate, as we have suggested, there should be corresponding shifts in range of the Arcto-Tertiary Flora in Eurasia. Tertiary floras are relatively rare in China, where studies were begun in 1925 and continued in 1933 and 1937. By contrast they are numerous in Japan, where field and museum work initiated in 1950 was greatly extended in 1951. There is now abundant evidence to indicate that the Arcto-Tertiary Flora, in which Metasequoia is the dominant conifer, occurs in northeastern Asia at latitudes progressively lower from Eocene to Miocene time, as is the case in North America and Europe. This around-the-world conformity in latitudinal position at successive stages seems to eliminate polar shift as a factor responsible for the occurrence of temperate forests in Greenland and Spitzbergen during the Eocene, as suggested by certain Europeans. For the assumption of such a shift in position of the North Pole, from the north Pacific to its present position since Eocene time, involves the neces-

sity of postulating a boreal climate in Alaska and Japan during the Eocene epoch, when the fossil record shows these areas were occupied by a rich temperate forest. The occurrence of similar vegetation in Eocene rocks on opposite sides of the Pacific and on the arctic islands north of Europe, and its southward shift in subsequent Tertiary time, also raise serious question as to the efficacy of continental drift as a causal factor in such latitudinal changes. In the light of evidence at hand in the northern hemisphere, it seems far more probable that forests have migrated in response to climatic changes than that continents have changed their position.

Living units of the Arcto-Tertiary Flora have been studied in such widely scattered areas as Iceland, Anatolia, and Kashmir. The distribution of these forests and their similarity in composition seem wholly consistent with a common origin to the north in early Tertiary time, though the possibility of a still earlier and different center of dispersal cannot now be dismissed. By contrast, marked dissimilarity between living forests of broad-leafed evergreens in southern Asia and Central America suggests a more remote relationship, a longer period of independent development. Comparison of Eocene floras, the Neotropical-Tertiary Flora from middle latitudes in North America and the Paleotropical-Tertiary Flora of Asia, likewise shows marked differences in composition and suggests that they were not derived from a common center as recently as Eocene time, as were the more uniform deciduous forests of the North. Relative paucity of fossil plants in the tropics may make difficult our discovery of the centers of origin of these Tropical-Tertiary Floras, but the problem is one for which the paleobotanical record must eventually provide the solution.

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DEPARTMENT OF EMBRYOLOGY

Baltimore, Maryland

GEORGE W. CORNER, Director

The year was one of active and uninterrupted work in the various lines of research established at the Department of Embryology, together with a good deal of exploratory study aimed at opening up pathways for future investigation. Two persons were added to the departmental staff, one of whom has already been associated with the Department for three years.

On July 1, 1951, Dr. Arpad I. Csapo, formerly Fellow, became a permanent member of the Department. A native of Szeged, Hungary, he studied medicine at the University of that city. After special training as an obstetrician, Dr. Csapo began research on the biochemistry of the muscular tissue of the uterus under the guidance of Professor Albert Szent-Györgyi. In 1948 he was invited to the University of Uppsala in Sweden, and in the next year came to the United States to work at the Department of Embryology. During the past two years Dr. Csapo has held an appointment as lecturer in obstetrics in Johns Hopkins University. The progress of his program of research has been recounted in these annual reports beginning with Year Book No. 49.

Dr. Bent G. Böving joined the Department in September 1951. After graduating from Swarthmore College, Dr. Böving spent one year as a graduate student in zoology under Professor Emil Witschi at the University of Iowa. He afterward took his M.D. degree at Jefferson Medical College, subsequently holding a fellowship of the U. S. Public Health Service (National Cancer Institute) at Yale University.

Dr. Chester H. Heuser, who retired on

August 31, 1950 after twenty-nine years of continuous service, was appointed a Research Associate. During the summer of 1951 he spent two months at the Department of Embryology working on human embryos in preparation for his proposed descriptive catalogue of the earlier stages (i to x) which were not included in the series "Developmental Horizons in Human Embryos" begun by the late Dr. George L. Streeter.

Dr. Fritz Fuchs and Dr. Anna-Riitta Fuchs, of Copenhagen, Denmark, concluded in December 1951 their year's work in the Department in association with Dr. Louis B. Flexner. Their stay was facilitated by grants from the American-Scandinavian Foundation and from the Carnegie Institution of Washington. The program of their work on the transmission of phosphorus across the placenta was outlined in Year Book No. 50; the results will be summarized when published.

Dr. Christine Gilbert, senior lecturer in anatomy at the University of the Witwatersrand, whose arrival for a year's stay as Fellow of the Carnegie Institution was mentioned in Year Book No. 50, completed her study of the early embryology of the baboon and returned home at the end of August 1951.

Mrs. Virginia B. Peters, Research Assistant to Dr. Louis B. Flexner under a special grant, completed her period of assignment to the Department of Embryology on June 30, 1952. During six years of association with the work of the laboratory, Mrs. Peters devoted herself with notable skill and thoughtfulness to the investiga-

tion of the microscopic structure of the central nervous system.

Dr. Ronald Singer, senior assistant professor of anatomy, the University of Cape Town, South Africa, arrived in November 1951 to spend about a year under a fellowship of the International Rotary Foundation. His program of research will be outlined below.

Dr. Harvey McKay Carey, post-graduate traveling fellow of the University of London, was in Baltimore from September 1951 to May 1952, dividing his time between the Department of Embryology and the Department of Obstetrics of Johns Hopkins University and Hospital. With the advice and guidance of Dr. S. R. M. Reynolds, Dr. Carey carried out investigations of the muscular contractility of the cervix uteri during labor, which will be described below under "Program of Investigations."

Dr. Anna W. Chacko, obstetrician and gynecologist of Peshawar, Pakistan, worked in the laboratory from October 1951 to June 1952 under the guidance of Dr. S. R. M. Reynolds, investigating problems which will be discussed below.

Dr. E. Carl Sensenig, of the University of Alabama School of Medicine, returned to the laboratory as in several previous summers, for a visit of about five weeks in August and September 1951, during which he studied the development of the human occipital and cervical vertebrae.

Dr. Theodore W. Torrey, chairman of the Department of Zoology at Indiana University, took advantage of his sabbatical leave to work at the Department of Embryology January to May 1952. The outcome of his intensive study of the development of the earliest stages of the embryonic kidney (pronephros) is mentioned below under "Program of Investigations."

Commander Roy E. Crowder, U. S.

Navy Medical Corps, in charge of obstetrics at St. Albans Naval Hospital, Long Island, New York, whose first visit to the laboratory was mentioned in Year Book No. 50, returned for two visits of about two weeks each, in November 1951 and May 1952, to continue his studies on the development of the human adrenal gland.

Miss Eleanor C. Adams, assistant to Dr. Arthur T. Hertig, Research Associate, at Harvard Medical School, spent the month of July 1951 at the Department learning the technical procedures for sectioning, staining, and reconstructing embryos, in preparation for assisting Dr. Hertig in writing the descriptions of the very early embryos mentioned in Year Book No. 50.

The following visitors spent a few days each at the Laboratory, making use of the Carnegie Collection of human and primate embryos, for study of the topics indicated after their names:

Dr. Alexander Barry, assistant professor of anatomy, University of Michigan: Development of the human heart in the first month. June 16–17, 1952.

Dr. Hans Elias, Chicago Medical School: Early development of the liver. December 31, 1951—January 3, 1952.

Dr. Philip G. Evans, Guy's Hospital, London, England, temporarily attached to the Department of Gynecology, Johns Hopkins Hospital: Development of congenital anomalies. October–November 1951.

Dr. James A. Miller, professor of anatomy, Emory University School of Dentistry: The notochord of human embryos in relation to the development of surrounding cartilage and of chordomas. June 23–27, 1952.

Dr. Catherine A. Neill, of London, England, in Baltimore as fellow in the Cardiac Clinic of Johns Hopkins Hospital: De-

velopment of the pulmonary artery and valve. August–September 1951.

Dr. William M. Rogers, assistant pro-

fessor of anatomy, Columbia University, New York: Early development of the thyroid gland. June 5–6, 1952.

PROGRAM OF INVESTIGATIONS

Morphology

Early human embryos. During his stay at the laboratory in July and August 1951 Dr. Chester H. Heuser began intensive work on the earliest embryos in the Carnegie Collection, that is to say those of horizons i to x of the Streeter classification, covering the age span from ovulation to 24 days. The collection now includes 65 specimens within this age span, a large proportion of which were obtained by Dr. Arthur T. Hertig and Dr. John Rock. About 35 of these are normal and well preserved and are therefore suitable for inclusion in the Horizons series which Dr. Heuser is preparing. As reported in recent Year Books, several extremely early embryos have recently been added. From the standpoint of the comparative embryologist a most valuable part of the series is that from 7 to 18 days, when the embryo is securing its attachment to the mother and when it passes through those primitive steps of organization which are common to all vertebrates, including the processes of gastrulation, formation of the amniotic duct, of the allantois, the notochord and chorda canal, etc. Upon such basal embryonic processes the biologist must depend as part of his evidence for determining evolutionary relationships in the animal kingdom. The relatively large number of specimens available in the Collection makes it possible to follow them in man more closely than before.

About 6 early embryos, cut by Dr. Heuser, were added to the available sectioned material. At the instance of Dr. G. W. Bartelmez, about 12 early human embryos, obtained and prepared by him

personally during his long connection with the University of Chicago, were donated by the Department of Anatomy of that institution. These embryos, some of which are of presomite age, constitute an important addition to the resources of the Department of Embryology. Several of them are superbly preserved.

Dr. Arthur T. Hertig, Research Associate, and Dr. John Rock have continued at the Free Hospital for Women, Brookline, Massachusetts, their search for early human embryos. Eleven uteri, obtained under circumstances suggesting the possible presence of early embryos, were examined but no embryos were found. This is the second year of the program (which has been going on for fourteen years) in which no early embryos have been recovered. During the whole span of the research, Hertig and Rock have found 36 early human embryos, all but one of which were less than 17 days of age. Thirteen were abnormal and 23 normal. Eight of them were in the preimplantation stage. Four of these are the only normal examples of this phase of human development yet discovered. other four showed various abnormalities, for example degenerating or multinuclear cells.

All eight of these extremely early embryos are described in detail in a manuscript which has been submitted for publication in the Carnegie Contributions to Embryology. From the histories of the individual cases, Dr. Hertig estimates the progress of development as follows: At 36 hours, 2-cell ovum, in the oviduct; at 72 hours, 12-cell morula, in the oviduct; at 96 hours, 58-cell blastula, in the uterus;

at 108 hours, 107-cell blastula, in the uterus.

Although the number of specimens is too small for statistically valid statements, the relative number of abnormal embryos in this series is higher before implantation (50 per cent of 4 cases) than after implantation (32 per cent of 28 cases). The normality or abnormality of the embryo seems to be determined by the intrinsic quality of the gametes (egg and sperm), inasmuch as the patients who produced the abnormal embryos were as fertile as those whose embryos were normal, and their reproductive organs revealed no greater degree of abnormality on clinical examination.

During the past year a histological and histochemical study of the corpora lutea from many of these early pregnancies, together with some from older gestations and others not associated with pregnancy, was published by Dr. Roger F. White in collaboration with Dr. Hertig, Dr. Rock, and Miss Eleanor C. Adams. This article is summarized below, under "Published Research."

Neural-crest cells of optic origin. Dr. George W. Bartelmez devoted much of his time during the year to investigation of an interesting discovery made by him some years ago at the University of Chicago in one of the embryos previously mentioned. His study of this specimen, in which he was assisted by Dr. Mary P. Blount, has now been extended to a large series of human and other primate embryos in the Carnegie Collection. In brief, the finding relates to a previously unnoticed source of cells of neural-crest type. In all vertebrates the developing brain and spinal cord, at a very early stage of development, before the neural groove is closed, give rise along their whole length to free cells which migrate outward and resemble embryonic connective tissue. These neural-crest cells participate in the formation of the sensory

ganglia which lie outside the central nervous system, and of the sheaths of the peripheral nerves. There is in addition good evidence that they produce various other structures such as the medulla of the adrenal gland. A very characteristic activity which has been demonstrated in all vertebrates that have been studied experimentally is the production of pigment cells. This has been seen not only in amphibians and birds, but also in the rat, in which the development of the neural crest is similar to that in man.

The optic vesicle is the first part of the eye to appear in development, and it is the center about which the other parts of the eye differentiate. Dr. Bartelmez finds that at the time when the optic vesicle is a simple fold of the future brain wall, it gives rise to cells of the neural-crest type in the same manner as other regions of the neural folds. At the stage of 15 to 25 somites (horizons xi and xii, about 23 to 26 days ovulation age), active migration of such cells from the wall of the optic vesicle is clearly evident in well prepared human embryos, and also, as supplementary observations reveal, in those of other primates, including the rhesus and cebus monkeys.

In view of the fact that the retina, which arises from the optic vesicle, acquires membranes similar to those of the brain and spinal cord, and these in turn have been shown in certain species to arise in part from neural-crest cells, the significance of Dr. Bartelmez' studies is obvious. In a report of the investigation, which will be published in volume 35 of the Contributions to Embryology, he suggests that in man the pigment cells of the uveal tunic (iris, ciliary body, and choroid) are derived from the neural-crest-like cells of the optic vesicle sheath which he has described.

Osteogenesis. Dr. Ronald Singer, Fellow of the International Rotary Foundation,

undertook at the suggestion of Dr. Bartelmez to study the time sequence of ossification in the embryo, examining for this purpose four typical bones, namely the mandible, clavicle, humerus, and femur. There is some indication that the two sexes differ in rate of ossification. One of the aims of Dr. Singer's study is to see whether this difference, if it exists, begins with the earliest differentiation of bone in the embryo. The Carnegie Collection is the only repository in the world of sufficiently numerous early embryos to make such a study possible. As usual in such research, numerous new or little-known facts have turned up. Among these is the observation by Dr. Singer that ossification of the mandible begins in the perichondrium of Meckel's cartilage, one of the few surviving bits of the primitive cartilaginous skeleton of the head (chondrocranium) that can be detected in the human embryo.

Dr. Singer has also taken up study of the development of deformities of the skeleton in "screw-tail" mice, a strain of animals having hereditary disturbances of bone growth, which Dr. E. Carleton MacDowell has been breeding at the Department of Genetics.

Pronephros. Dr. Theodore W. Torrey, of Indiana University, made a comprehensive survey of the development of the earliest rudiments of the human kidney, i.e. the pronephros and its paired ducts, in all available embryos of relevant age in the Collection. Details of the pronephros in single embryos have been described in many monographs in the Carnegie Contributions. Dr. Torrey has gathered these together, comparing and elucidating them. The subject has been very obscure, as is well known to puzzled readers of the chapter on the urinogenital system in the Keibel-Mall Handbook of Human Embryology. Dr. Torrey's description and discussion, which will appear in volume 35 of the Contributions to Embryology, clear away a good deal of the confusion which has been due to insufficiently clear comparison of the conditions in the human embryo with the general vertebrate pattern, and to over-schematic distinction of the pronephric from the mesonephric series of renal primordia. Supposed peculiarities of the pronephros in man are seen not to exist, and therefore in this as in many other details of embryonic structure our species is found to be more like other vertebrates than has sometimes been supposed.

Cerebral veins. Mrs. Dorcas Hager Padget almost completed during the year the draft of a summary statement of the development of the cerebral veins in man. Illustrations for this report, which are necessarily very detailed and complicated, are being prepared by the investigator, who is a trained medical illustrator.

EXPERIMENTAL BIOLOGY

Urinogenital system. Dr. Robert K. Burns continued his studies of the general morphology of the developing testis and associated structures in the opossum embryo, as a basis for comparison with specimens in which the development of the male reproductive tract was modified by experimental treatment with hormones during early embryonic life.

In February and March 1952 Dr. Burns spent about six weeks at the field station of the University of Florida at Welaka, trapping opossums and carrying out experimental procedures on the pouch young. A considerable addition to his study material was thus made. The Department of Embryology is deeply indebted to the Department of Biology at the University of Florida for generous provision of laboratory space and other facilities.

Implantation of the embryo. Dr. Bent G. Böving conducted experimental observa-

tions on the implantation of the rabbit embryo in the uterus. With the aid of apparatus which he invented and brought with him to the laboratory for this purpose, improving and modifying it as the work proceeded, he was able to observe the living blastocysts in the uterus and to study certain phases of their positioning in readiness for attachment and of the process of attachment itself. An interesting discovery made during these observations was that a restricted area of the blastocyst wall, at the side opposite the embryonic area, develops a well marked alkaline reaction, and the zona pellucida overlying that region becomes soft, sticky, and weak. The implanting blastocyst, as observed in the Böving plastic chamber apparatus, is held close to the antimesometrial wall of the uterus, which relaxes locally to form a domelike space. In this space the blastocyst is kept moving to and fro and rotationally until finally the altered area of the zona pellucida is held in prolonged contact with the uterine lining, and attachment occurs. A motion picture film prepared by Dr. Böving showing these events with explanatory diagrams was received with much interest at the 1952 meeting of the American Association of Anatomists. Dr. Böving is continuing his observations with the aim of elucidating the mechanisms of attachment, orientation, and implantation of the mammalian embryo. His fundamental interest is in the mechanisms of tissue invasion such as are seen in various growth processes and in tumors. The implanting embryonic tissues offer a striking example of such a mechanism which is accessible to observation and experiment.

Placental vessels. Dr. Elizabeth M. Ramsey was able during the year to bring to a successful conclusion her investigation, mentioned in Year Book No. 50, of the venous drainage of the placenta in the rhesus monkey. The widely quoted views

of Spanner on the subject of the blood circulation in the human placenta include very definite statements about the pathway of drainage of placental blood into the maternal veins. Dr. Ramsey has investigated this same question in a series of 23 monkey placentas of known age, in which the blood vessels were injected with ink. An illustrated account of the results is being prepared for volume 35 of the Contributions to Embryology. Dr. Ramsey plans to extend her studies to the human placenta and already possesses a number of successfully injected specimens.

FETAL CIRCULATION

In September 1951 Dr. S. R. M. Reynolds returned from England, where he had spent a year on a Guggenheim Fellowship, working at the Nuffield Institute for Medical Research at Oxford. The program and results of his research on the fetal circulation by cine-radiography, done in collaboration with Drs. Gordon M. Ardran, M. M. L. Prichard, Peter Daniel, G. W. Dawes, and Derek Wyatt, of the Nuffield Institute, were fully outlined in Year Book No. 50. During the ten months following his return, Dr. Reynolds has been engaged in preparing definitive reports for publication (which will be summarized in these Year Books as they appear), and in planning for continuation of the program. He has also begun assembling oscillographic and other apparatus for study of the development and control of the fetal circulation.

Certain special problems arising from or re-emphasized by the cine-radiographic studies at Oxford were investigated by Dr. Reynolds and co-workers in Baltimore after his return. A summary of these follows:

Lung circulation. X-ray motion pictures of the lungs of sheep fetuses at term

showed that immediately after the establishment of respiration there is a very great increase in the amount of blood flowing through the lungs. Presumably this means that the terminal blood vessels (alveolar capillaries) suddenly become distended. To gain direct evidence on this point, Dr. Reynolds with Dr. Kenneth Rhaney, of the Johns Hopkins Department of Anatomy, observed the lungs of living guinea pigs at term, before and after the onset of respiration, injecting a fluorescent dyestuff into the circulating blood to visualize the pulmonary circulation. A striking difference was apparent, for in the fetal lungs there was only minimal blood flow about the pulmonary alveoli, whereas immediately after the newborn animal began to breathe there was a copious amount of blood in dilated capillaries about the alveoli.

Ductus venosus. Dr. Anna W. Chacko. working under the direction of Dr. S. R. M. Reynolds, studied the development of a localized constriction in the ductus venosus of the embryonic liver. Reasons for believing that this little-known structure, acting as a sphincter, participates in the control of blood flow in the umbilical vein are cited in an article by Dr. Reynolds in the Scientific American for July 1952. Dr. Chacko has identified this constriction ring when it first appears in embryonic life, has traced its further development, and has shown that it is well established by the early fetal period. A description is being prepared for publication.

Architecture of the umbilical vessels. Dr. Chacko, under the direction of Dr. Reynolds, has also studied the structure of the umbilical arteries and veins, comparing them in the distended and constricted states respectively. These particular vessels were chosen primarily because of their cardinal importance in the phenomena of

fetal circulation, but as it happens they are also very favorable for study from the standpoint of general histology, because their isolated situation in the cord makes them easily observable apart from other tissues, and they can readily be studied in long segments and subjected to experimental procedures. Dr. Chacko is preparing a description of the vessel walls, in which the muscle is arranged in interlacing spirals, primarily to resist lateral distention. and the elastic tissue chiefly in longitudinal laminae, to resist lengthwise extension. It has been found that constriction of the umbilical arteries is associated with shortening. Thus, both the radial and axial dimensions of the artery are reduced by constriction, its walls undergo thickening, and consequently the volume of the lumen is reduced. A relatively slight constriction is sufficient practically to occlude the artery.

The work includes a study of "Wharton's jelly," the connective tissue of the umbilical cord, which Dr. Reynolds and Dr. Chacko regard as a modified adventitial layer of the cord vessels. In the functional umbilical cord, in which the vessels are fully distended, the Wharton's jelly is thinned out into almost imperceptible interstitial sheaths. Only when the blood vessels have shrunk does it appear as a bulky, loose jelly-like tissue, such as is illustrated in textbooks.

Force of Uterine Contractions

Dr. Harvey M. Carey and Dr. Reynolds have worked together on apparatus and techniques which Dr. Carey has applied to patients in the Johns Hopkins Obstetrical Clinic, by which the pressures set up within the fetal membranes by the force of uterine contraction are measured and recorded. The method, suggested by Dr. Reynolds, makes use of a tube inserted aseptically high into the amniotic cavity

and connected to a strain-gauge pressure recorder. Calculations from records obtained in this way show that effective dilatation of the cervix in labor depends upon the attainment of a critical pressure acting upon the uterine contents. In the light of this basic concept, the work has been extended to include evaluation of the effects of sedative drugs on the effectiveness of labor contractions.

BIOCHEMISTRY AND PHYSIOLOGY OF UTERINE MUSCLE

In the two previous Year Books (Nos. 49 and 50) mention has been made of an intensive investigation of the fundamental characteristics of the muscular tissue of the uterus. Dr. Arpad Csapo's investigations in Hungary and Sweden, using modifications of the techniques worked out by the Szent-Györgyi school in studies on skeletal muscle, showed that uterine muscle contains the same contractile proteins as other muscular tissues. The uterus, however, is known to be controlled in a highly specific way by the ovarian hormones, contracting with quite different force and rate in diverse states of the reproductive system, such as estrus and pseudopregnancy. Inasmuch as Dr. S. R. M. Reynolds and Dr. George W. Corner have acquired considerable experience in the endocrine aspects of this problem, it was thought that a combined attack by biochemical and physiological methods, to be conducted by Csapo in consultation and collaboration with Reynolds and Corner, might extend our knowledge not only of uterine muscle contraction, but of the general phenomena of the effects of hormones on the uterus. This effort is now in its fourth year.

The principal activity in the year under report was the study of the contractile power of uterine muscle when stimulated to contract isometrically; that is to say,

when it is made to deliver all its force in pulling rather than in merely shortening. For this study, apparatus was assembled by Dr. Corner, with which, for the first time so far as is known, isometric contractions of uterine muscle elicited by electrical stimulation have been quantitatively recorded. With this method, which gives a more precise measure of contractile force than the isotonic recording formerly used, it was discovered that uterine muscle, like that of the heart, exhibits the "staircase phenomenon"; that is, the tension it exerts at a single contraction depends upon the length of time since the previous contraction. Within certain limits, the longer the interval, the less the tension developed. Using castrate rabbits injected with the two ovarian hormones, it was then found that the characteristic slope of the staircase of uterine muscle under the influence of estrogen is reversed under progesterone domination. It is evident therefore that the two hormones have opposing effects on some factor or factors intimately involved in the mechanism of contraction. A hint, already found by Szent-Györgyi and Hajdu in their work on heart muscle, that the staircase phenomenon is related to the concentration of potassium in the muscle cells is being actively followed up in the experiments on uterine muscle, which offers an unusually favorable opportunity for such an investigation because its responsiveness can be experimentally controlled by hormone treatment.

This controllability of the uterine muscle tissue has also been used in a study, by Csapo and Corner, of the relation between the amount of tension which the muscle develops and the concentration of the contractile protein, actomyosin, present in it. Csapo had previously shown that the amount of actomyosin can be varied by hormone treatment; it drops practically to zero after castration and is restored by in-

jection of estrogenic hormone. Using a series of castrate animals given graded estrogen treatment, the gain in tension was found to be approximately parallel to the increase in actomyosin. This finding strongly supports the view that actomyosin is the substance basically responsible for the phenomenon of muscular contraction.

Mr. John H. Menkes, of Johns Hopkins Medical School, and Dr. Csapo have studied the effect of the ovarian hormones upon the final energy-supplying constituents of uterine muscle, namely the adenosine nucleotides (adenosine tri-, di-, and monophosphate). Using, with the advice of Dr. Beni Horvath, the recently introduced Kalckar enzyme technique, it was found that estrogen treatment increases the concentration of all three of these compounds in the uteri of ovariectomized animals. Thus the ability of the uterine muscle to build up both the contractile protein and the energy stores for the work of contraction is dependent upon the estrogenic hormone. The investigation just summarized, of the three adenosine phosphate compounds, is an extension of previous work by Menkes and Csapo discussed below under "Published Research."

Work on other aspects of this complex problem is in less advanced stages. Dr. Csapo has accumulated a large number of observations on the energetics of uterine muscle strips, under full oxygenation, in complete anoxia, and with aerobic and anaerobic glycolysis inhibited by enzyme poisons. These data have not yet been subjected to calculation. At Woods Hole in the summer of 1951 Dr. Csapo studied the fate of adenosine triphosphate in the blood, finding that in a blood sample containing ATP the hemolyzing red blood cells release an enzyme (ATP-ase) which

destroys the ATP. He also continued investigation of a peculiar "x factor" which alters the contraction of actomyosin threads and which he had found to affect the recombination of actin and myosin. The unknown factor turned out to be myokinase, the enzyme which catalyzes the reaction $2 \text{ ADP} \rightarrow 1 \text{ ADP} \rightarrow 1 \text{ AMP}$. In the absence of this enzyme ADP accumulates, inhibiting the recombination of actin and myosin.

In consultation with Dr. Csapo and with the technical advice of Dr. Bartelmez, Mr. George W. Settle, of Johns Hopkins Medical School, began an investigation of the size of uterine muscle cells as conditioned by the ovarian hormones.

The results of long-continued research programs of this kind may not seem fully coherent when set forth, as in these Year Books, partly as a report on work in progress and partly as abstracts of published articles on detailed aspects of the work. A systematic review of progress up to the summer of 1952 will be presented in the Ciba Foundation Lecture by Dr. Csapo and Dr. Corner, to be delivered by Dr. Corner in London in December 1952. The lecture will probably be in print by the time this Year Book is ready for circulation.

BLOOD SUBSTITUTES

After the departure of Dr. Louis B. Flexner for his new post, Dr. Francis P. Chinard took full charge of the experimental studies on the permeability of blood vessels with respect to certain blood substitutes. This investigation was undertaken at the request of the National Research Council, under sponsorship of the U. S. Army. Progress has been made but the results are not ready for reporting here.

CO-OPERATIVE RESEARCH ACTIVITIES

The group conducting a study of gestation in relation to general health, at the Columbia-Presbyterian Medical Center, New York City, under the direction of Dr. Katharine K. Merritt, to which reference was made in Year Books Nos. 49 and 50, has terminated its collection of data. The co-operation afforded by the Department of Embryology for the past three years, by examining and reporting upon early abortions, has come to an end. In a letter acknowledging the Department's services, Dr. Merritt commented in particular upon the excellence of the informa-

tion supplied by Dr. Elizabeth M. Ramsey.

Co-operation continues with the Department of Gynecology of Johns Hopkins Medical School by the provision of facilities, assistance, and the care of monkeys for research on experimental production and treatment of endometriosis. Laboratory facilities and technical assistance were furnished to the Departments of Biochemistry and of Medicine of Johns Hopkins University for the use of Dr. Francis P. Chinard in developing a colorimetric method for assaying the blood substitute polyvinylpyrrolidone in body fluids.

PUBLISHED RESEARCH

HORIZONS OF HUMAN DEVELOPMENT

When Dr. George L. Streeter died in 1948, he left unfinished the last five stages of his great series, "Developmental Horizons in Human Embryos." All that was available, with which his colleagues could complete the unfinished portion, was the illustrations and some bundles of notes. Dr. Chester H. Heuser, aided by Dr. Corner, has painstakingly organized and expanded these notes into an article of about the same size and almost the same degree of completeness as those which Dr. Streeter published. The period of development from Streeter's stage xi (23 to 25 days) to the end of the embryonic period, his stage xxiii (46 to 48 days), has now been entirely covered.

As each installment of this series was printed, 500 copies were kept unbound in sheets. With the completion of horizons xi to xxiii the sheets were bound up into a special volume and provided with an explanatory preface by Dr. Corner. This book, entitled *Developmental Horizons in Human Embryos: Age Groups XI to*

XXIII, and designated as Embryology Reprint Volume II, is available through the Office of Publications of the Carnegie Institution of Washington.

GROWTH OF THE SPINAL CORD

Volume 34 of the Contributions to Embryology includes an account of the dimensional growth of the spinal cord in the fetal and infant rhesus monkey, by Dr. Marion Hines and Dr. Bettina M. Emerson. The former, now professor of experimental anatomy at Emory University, was associate professor of anatomy at Johns Hopkins Medical School, and the latter a student of medicine there, when the research was in progress. Most of the 22 specimens studied were bred by Dr. Carl G. Hartman in the monkey colony of the Department of Embryology. The authors have painstakingly measured and compared the length of the spinal cord and its various subdivisions, and have worked out the relative changes of these measurements as growth proceeds. The areas of cross sections of the cord, the areas of the

gray and white matter, and the areas of the three funiculi of the white matter were similarly studied. The investigation covers the period from the 9th week of gestation to 14 months after birth. A 5-year-old adult monkey was also studied for comparison. The major importance of this work lies in the fact that the physiological development of each of the fetuses and infants had been exhaustively studied during life, and the anatomical differentiation within the spinal cord could therefore be interpreted in terms of known physiological status. Two previous reports on the same animals have been published in the Contributions to Embryology, namely, Hines and Boynton on the maturation of excitability in the precentral gyrus (vol. 28, 1940) and Hines on the development of reflexes, postures, and progression (vol. 30, 1942). Much of the information given in the present paper is too detailed for consideration here. Among the interesting general findings are the following: (1) The increase in size of the spinal cord and of its constituent parts between birth and maturity (5 years) is comparable with that which takes place in man in his longer span between birth and maturity. (2) The relation between growth in length of the cord and the cross-sectional area at various levels does not follow J. Huxley's general formula for the relation between any two dimensions of a growing organism (y= bx^k) except in the lower thoracic region, which is the least differentiated part of the spinal cord. Greater differentiation is accompanied by deviation from this theoretical relation. (3) The greatest increment in area of all the morphological entities measured, and in length of all subdivisions of the cord except the lumbar, occurs between 100 and 125 days of gestation, preceding establishment of the most fundamental reflex pattern, that of respiration. The greatest augmentation of growth in

length of the lumbar part of the cord, however, occurs between the end of the first year of postnatal life and adolescence, and is thought to represent the period of differentiation necessary for functional activities related to sexual maturation. (4) Mathematical relations between the increments of the various entities measured suggest that the size of each part of the spinal cord at a given stage of growth is regulated in two ways: (a) as a proportion of the ultimate dimension of the individual entity attained in the adult of the species, and (b) as a proportion of the dimensions of the whole cord at the given time in development.

DEVELOPMENT OF THE MENINGES

As part of a series of investigations concerned with the development of the vertebral canal and its contents, Dr. E. Carl Sensenig, of the University of Alabama, has studied the early development of the meninges of the spinal cord in human embryos in the Carnegie Collection. Seventyfive specimens were examined, from embryos of horizon x (20-22 days gestation age) to fetuses of 12 weeks. The chief question at issue among comparative embryologists with respect to the meninges is whether these membranes surrounding the brain and spinal cord are derived entirely from mesoderm, or whether (as some investigators of lower vertebrates have stated) the leptomeninx (pia mater and arachnoid) is largely derived from neural-crest cells of ectodermal origin. Dr. Sensenig, while agreeing that in human embryos neural-crest cells mingle with the undifferentiated mesenchyme that surrounds the early neural tube, finds that the intermingling is so complete that the contributions of the two germ-layer elements to the developing pia mater cannot be distinguished. The arachnoid portion of the leptomeninx forms by delamination from the inner surface of the dura mater. The evidence thus supports a mesodermal origin for all the meninges, with the reservation that ectodermal cells from the neural crest participate in the early formation of the tissue that gives rise to the pia mater. The subsequent differentiation of the three layers into the definitive pia mater, arachnoid, and dura mater, and of the subdural and epidural spaces, is described and illustrated in detail in Dr. Sensenig's article.

ORIGIN OF THE MÜLLERIAN GROOVE

Very little information has been available concerning the exact site of origin of the human Müllerian or paramesonephric duct and its exact relation to the other elements of the early urinogenital system. Dr. Robert J. Faulconer has investigated this question in 7 human embryos of relevant age in the Carnegie Collection. He finds that the site of invagination of the Müllerian groove varies greatly in its relation to the surface of the mesonephros (Wolffian body) and may occur on almost any part of the ventral and lateral surfaces of that organ. Its location may be different on the two sides of the same embryo. The site of invagination is an area of specialized coelomic epithelium which is thought to represent one or more mesonephric nephrostomes.

HEAD CAVITIES IN HUMAN EMBRYOS

The contents of Dr. Perry W. Gilbert's paper on this topic, which is listed in the appended bibliography, were fully summarized in Year Book No. 50 (p. 143).

SEX TRANSFORMATION IN THE OPOSSUM

The report given by Dr. Robert K. Burns at a colloquium on sex differentiation in vertebrates, held in Paris at the Collège de France in June 1950, has been published.

Summarizing much of Dr. Burns' work during several years past, this report deals first with his general conclusions from experiments on the control of sex differentiation in the embryonic (pouch young) opossum by sex-gland hormones. The exact way in which male and female structures are induced to form from the indifferent embryonic primordia has been subject to varying interpretations. The central problem is whether male (testicular) hormones determine the differentiation of male structures and female (ovarian) hormones determine the differentiation of female structures by direct and sex-specific action upon primordia, or whether, on the contrary, there are indirect complicating effects such as suppression, by a given hormone, of the directive influence of the gonad which normally causes the affected organ to assume the characteristics of one sex or the other. The problem has been further complicated by experiments in which large doses of a given hormone have caused the undifferentiated sex ducts of both sexes to develop simultaneously; for example, testicular hormone causes both the potentially male and potentially female primordia to develop. This "paradoxical" effect never occurs with low dosages in what may be considered the physiological range.

As a result of his long and careful studies Dr. Burns considers that his results in the opossum are generally compatible with what may be called the classic conception of hormone action, according to which each type of hormone acts selectively upon the appropriate sex primordia, stimulating or inhibiting according to their inherent capacities. His results with injected hormones support the view that the embryonic gonads produce hormones which control differentiation of the accessory sex organs, and that these hormones are probably similar in chemical nature to those which are produced by the mature testis and ovary.

In the second part of his paper Dr. Burns presents the details of experiments, referred to in Year Book No. 49, in which the female hormone estradiol dipropionate caused changes in the embryonic testis, including survival of the germinal epithelium and the development therefrom of a cortical zone containing cordlike invaginations; that is to say, the testis was modified in such a way as to acquire the characteristics of an ovary. This paper, as one of the European participants said in discussing it, marks an epoch in the study of experimental sex differentiation in mammals, for it is the first report of the production of an indisputably intersexual state in a mammalian gonad by treatment with crystalline hormones. The failure of previous experimenters, and of Dr. Burns himself in earlier experiments, to obtain such an effect is explained by the facts that treatment must be begun before the germinal epithelium of the testis disappears (i.e. during the first 10 days of pouch life), and must be continued beyond 20 days in order to include the period of active cortical differentiation. Previous experiments satisfied one or the other of these conditions, but failed to combine the two in a single experiment.

SEX DEVELOPMENT IN THE HAMSTER

Dr. Faith Wilson LaVelle has carried out a comprehensive investigation of the development of the reproductive tract of the golden hamster. This animal has the practical advantages, for experimental use in problems of the reproductive system, of large and frequent litters and a very short gestation period, 15 to 16 days. Dr. La-Velle's primary aim was to investigate the relation between gonads and reproductive tract during sex differentiation, and to determine the extent to which the adrenal

cortex may participate in that process through hormonal action. Inasmuch as very little was known until recently about the early development of the hamster, Dr. LaVelle's description of the normal structure of the reproductive system will be of general interest to comparative embryologists, as will her account of the development and growth rate of the various constituent organs. Her experimental findings include the fact that removal of the gonad affects the growth and differentiation of the reproductive tract in the male much more rapidly and profoundly than in the female. It is concluded that the testis of the hamster influences the growth of the accessory reproductive organs from birth onward, whereas the ovary has no significant effect until the time of puberty. The reproductive tracts of both sexes are capable of early response to treatment with androgenic hormone. The dorsolateral pigment spots of the adult hamster show a characteristic difference in the two sexes, coarse black hairs appearing normally in males only. Dr. LaVelle has shown that the male pattern depends upon androgen action.

The adrenal cortex of the young male hamster normally lacks the special condensed juxtamedullary zone found in the female after 27 days. After castration the zone develops in the male, and in the female it persists. Its appearance under such circumstances in the male may indicate compensatory adrenal activity. No evidence of such activity could, however, be obtained. Pituitary adrenocorticotrophin does not delay the appearance of castration effects. On the other hand, the same hormone given to young noncastrate animals produces precocious development of the prostate, seminal vesicles, and bulbourethral glands.

THE HUMAN CORPUS LUTEUM

Dr. Arthur T. Hertig, Research Associate, and Dr. John Rock, whose indefatigable efforts in collecting human embryos have been recorded annually in these reports, have had opportunities to gather together a large number of ovarian biopsy specimens. The corpus luteum of pregnancy has been obtained with practically all the early embryos of the Hertig-Rock series. Forty-one corpora lutea ranging from 2 days to 41/2 months in age have been subjected to intensive cytological and histological study, conducted chiefly by Dr. Roger F. White in consultation with his coauthors Dr. Hertig, Dr. Rock, and Miss Eleanor C. Adams. From the detailed description the following points are chosen for mention because of their general interest to students of reproduction: (1) Before implantation of the ovum, no difference can be discerned between chronologically similar corpora lutea of the normal menstrual cycle and those associated with normal or abnormal ova. (2) After implantation, at about 6 or 7 days after ovulation, the corpus luteum does not undergo regression, but is stimulated to increasingly higher levels of functional activity (as assumed from the microscopic structure), up to 6 weeks of menstrual age (i.e. approximately 56 days ovulation age). (3) As the mature ovarian follicle nears ovulation, a number of distinctive cells ("K cells") become conspicuous in the granulosa lutein layer and subsequently enter into formation of the corpus luteum. These are thought to constitute a special cell type which is the site of intense localization of ketonic lipids. (4) When the implanted ovum is deficient in trophoblastic development, the corpus luteum undergoes early regression. The most striking feature of such failure of the corpus luteum is colloid degeneration of the "K cells."

Cyclic Changes in the Endometrium of the Rhesus Monkey

A paper by Dr. G. W. Bartelmez represents the completion of a project formulated by Dr. Carl G. Hartman, Dr. Bartelmez, and Dr. George W. Corner almost twenty years ago, under which the cyclic changes of the ovary and uterus of the rhesus monkey were to be described from material collected by them at the Carnegie Department of Embryology, the University of Chicago, and the University of Rochester, augmented by specimens made available by Dr. H. B. Van Dyke from his work at Peiping Union Medical College in China. Of all the animals other than man which exhibit the phenomenon of menstruation, the rhesus monkey is the most available for study, and practically speaking is the only one which is available in numbers sufficient for experimental investigation. With the addition of Dr. Bartelmez' thorough and intensive description of the uterus, it may now be said that more is known about the cyclic changes in the reproductive system of rhesus than of any other primate. In man, for example, the endometrium associated with a fully mature follicle in the ovary has been described in only one case; the Bartelmez-Corner-Hartman-Van Dyke monkey material includes eight such cases. The study is based on 197 well preserved and well documented specimens, 129 of which were selected for presentation. Dr. Bartelmez recognizes follicular, progravid, regressive, menstrual, and repair phases. The descriptive criteria involve matters of technical detail which cannot profitably be summarized here.

Signs of regression appear after the 15th day of pregnancy, with ischemia and even

slight local hemorrhage ("placental sign" bleeding). In the absence of pregnancy this ischemia progresses to the stage of involution and menstrual bleeding. The rate and degree of premenstrual involution vary so greatly that anovulatory menstruation cannot be distinguished from ovulatory types with extreme premenstrual involution.

The cyclic changes are interpreted as specific adaptations: the follicular phase for insemination; the progravid reaction for the care of the implanting blastocyst; the regressive phase (which includes early placental bleeding) as providing pabulum for the invading trophoblast. Dr. Bartelmez emphasizes particularly the variability of the response of the endometrium to the ovarian hormones. This article, because of the wealth of material, the high degree of physiological correlation with the known reproductive status of the individual animals, and the author's cautious analysis, cannot fail to provide both stimulus and critical guidance to students of the physiology of reproduction as well as to gynecological pathologists and others who are concerned with the dating and diagnosis of human endometria.

Mitosis in the endometrium. At the suggestion of Dr. Bartelmez, Miss C. M. Bensley, of the University of Chicago, studied the rate of mitotic division of uterine epithelium in the rhesus monkey, using 50 cases, both pregnant and nonpregnant, selected from those mentioned above. The laboriousness of this work is indicated by the number of cells closely studied, 200,812. The material was divided into eleven groups representing functional states, chiefly on the basis of ovarian findings. The peak of mitotic activity is centered about the time of ovulation. Mitotic division of the epithelial cells continues during the luteal phase, but practically ceases during the late luteal and ischemic

phases. The facts thus ascertained form a part of the criteria utilized by Dr. Bartelmez in his general study cited above. One of the most significant facts revealed by the work is that cell division after the menstrual destruction has occurred is practically limited to the superficial glands. The epithelium of the deeper glands, which has generally been credited with the restoration of the whole mucosa, shows very little mitotic activity during the repair phase.

PREGNANCY IN THE BABOON

Mention was made in Year Book No. 50 of a study of the early embryology of the baboon, Papio ursinus. This research (which will be reported by Dr. Christine Gilbert and Dr. C. H. Heuser in volume 35 of the Contributions to Embryology) resulted from a co-operative enterprise under the direction of Dr. Joseph Gillman, Research Associate of the Carnegie Institution, at the University of the Witwatersrand, Johannesburg, South Africa. Financial support which enabled Dr. Gillman to initiate long-term observation and experiments on the development and gestation of the baboon was provided by grants from the Carnegie Institution through the Department of Embryology. Dr. Gilbert's stay of a year at the laboratory as Fellow of the Carnegie Institution of Washington is mentioned above.

Another valuable report resulting from this work has recently been published in South Africa by Drs. Gilbert and Gillman. This is an account of pregnancy in the baboon. Very little exact information has been available on gestation in primates other than man, chimpanzee, and the rhesus monkey. Gilbert and Gillman followed 14 female baboons throughout pregnancy, and in addition studied 5 which were pregnant when received and 6 mated for the purpose of obtaining the early

embryos now in the Carnegie Collection at Baltimore. Some of the detailed findings may be briefly summarized. The length of gestation was 173 to 193 days, averaging 187. Twelve of the 14 births occurred between days 184 and 193. The sexes of the offspring were equally distributed. Matings occurred at all seasons of the year, but 10 of the 14 births occurred in the winter and spring months. During pregnancy there was suppression of the rhythmical fluctuations in perineal swelling characteristic of this species; but perianal tumescence persisted. Profound changes in the sex-skin color occurred beginning 7 days after the first missed menstrual period. Lactational amenorrhea usually lasted 6 to 8 months. At birth the babies weighed 500 to 1212 grams, averaging 861 grams. The average weight of the males was 150 grams greater than that of the females. Many other facts of interest to students of gestation and fetal growth are set forth in the article.

FORM AND SIZE OF THE PREGNANT HUMAN UTERUS

One of the important features of the accommodation of the pregnant uterus to its growing contents (i.e. the infant, placenta, membranes, and fluid) is that the uterus undergoes a characteristic pattern of changing shape during its enlargement as gestation proceeds. These changes in shape have been stressed in a number of studies and communications by Dr.S.R.M. Reynolds and his co-workers, which have been reviewed in Year Books Nos. 46, 47, 48, and 49. In 1948 Dr. Reynolds presented to a group of physicians at the Johns Hopkins Hospital the results of his study of uterine accommodation in the rabbit. One of his hearers, Dr. J. T. Baker, an active obstetrician of Easton, Maryland, began to

incorporate in his regular office examination of maternity cases biweekly observations on the height and width of the uterus. The data thus obtained from 50 patients have been analyzed by Dr. Reynolds and Dr. Baker with respect to the relation of parity (i.e. the number of children born previous to and including the pregnancy studied) to the pattern of uterine enlargement. The results, as summarized by the authors, are as follows: (1) The uterus enlarges proportionately in three dimensions until the phase of relative elongation or conversion begins, in multigravidae at the 18th week, in primigravidae in the 21st week. (2) The fundus reaches the same height, on the average, in primi- and multigravid women, but the uterus attains its ultimate height somewhat faster in primigravidae. (3) The multigravid uterus is wider at term, but the primigravid uterus attains its ultimate width more rapidly. The uterus of the primigravid woman elongates more readily than it widens, in contrast with the multigravid. (4) At term the length exceeds the width in the primigravid uterus by 45 per cent; in the multigravid by 36 per cent. (5) If an index of elongation is calculated by dividing the length of the uterus by the width, its value regresses steadily after conversion; that is to say, the proportion of the relative elongation vet to be achieved diminishes with time. A mathematical formula for the rate of regression is given. The rate is faster in multigravidae than in primigravidae.

BLOOD PRESSURE IN THE UMBILICAL CORD

The quantitative aspects of blood flow in the umbilical cord have not been recorded by modern methods. From observations on sheep and goats something is known about the approximate mean pressures in the umbilical arteries and veins as measured by inserting a cannula connected to a manometer. Similar observations have been made on human umbilical cords, usually at the time of labor or under the difficult circumstances of an aseptic operation on the pregnant uterus. Dr. S. R. M. Reynolds has attacked this problem with equipment of his own design, using strain-gauge pressure transducers. Observations were made on 8 sheep especially bred for the purpose, and studied at stages of pregnancy ranging from 100 to 114 days. (The length of gestation in the ewe is about 147 days.) In 5 of the 8 experiments the very numerous and serious difficulties were overcome, so that extended and repeated observations were made on the fetal heart rate, arterial and venous pressure levels and gradients along the cord, the effects of maternal hypercapnia, and the effects of partial and complete occlusion of the umbilical blood vessels. These results, which are of importance in working out the physical and mechanical aspects of umbilical blood flow, need not be summarized quantitatively here. Dr. Reynolds deduces from them that the return of blood from the placenta to the fetus is brought about by several factors. One of these is an arrangement of the vessels in the cord such that they are able to act as a pistonless pump or pulsometer. Another is a difference in pressure between the placental end of the umbilical vein and the inferior vena cava of the fetus, and still another is a sphincter valve at the point of entrance of the venous blood into the ductus venosus, which maintains a high pressure in the umbilical vein and keeps it distended. As mentioned above, Dr. Anna W. Chacko has investigated the structure and embryological development of this sphincter.

Do Pulsating Arteries Dilate?

Anyone who has ever felt the human pulse at the wrist will have taken it for granted that the throbbing he feels is the result of alternating dilatation and contraction of the artery. During the course of experimental observations on the sheep's umbilical cord by use of X-ray motion pictures, Dr. Reynolds and his co-workers at the Nuffield Institute, Oxford, obtained pictures of the pulsating umbilical arteries, which were clearly defined because a substance opaque to X rays (Thorotrast) was flowing in the blood stream. Careful measurements of these pictures show that there is no lateral enlargement (dilatation) of the artery as the pulse passes through it. The whole artery is, however, displaced laterally, especially in the vicinity of curves where there is deflection of streamline flow, just as a hose is caused to whip about by a strong flow of water through it. The fact that a pulsating artery does not dilate was also found to hold for a large artery within the body, by X-ray studies on cats which showed that although the upper aorta (ascending and arch portions) is greatly expanded by blood periodically ejected from the heart, the descending aorta does not measurably dilate. The pulse wave is transmitted as a wave of pressure within the moving blood stream, not as a traveling bulge in the arterial wall. What is felt when the fingers are placed over an artery is the pressure pulse in the flowing blood, and also, at times, the lateral movement of the whole artery.

FACTORS IN DILATATION OF THE CERVIX

In lectures given in 1951 at London and Brussels, Dr. Reynolds discussed the physiological mechanisms by which the cervix uteri is dilated in labor. He pointed out three fundamental elements in this process.

The first of these is morphological; that is, the muscular tissue of the uterus at term is so disposed as to give a preponderance of contractile power to the fundus, and thus the contractions of the organ as a whole result in yielding of the cervix. The second element is physical, namely a set of relations between the form of the contracting uterus, as expressed by the principal radii of curvature, and the tension developed in the various portions of the wall of the organ. These relations also favor the fundus. The third element is biochemical. Csapo has shown that the concentration of actomyosin, the contractile protein complex of uterine muscle, is greater in the fundus uteri at term than in the corpus and cervix. When these elements are properly co-ordinated, a rhythmic pattern of contraction results by which the dominant fundus overcomes the resistance of the cervix and dilatation occurs, thus permitting delivery of the infant.

UTERINE MOTILITY IN MISSED ABORTION

A brief paper by Dr. G. Rowe-Dutton, Dr. Samuel Lubin, and Dr. Richard Waltman (all of the Cumberland Hospital, Brooklyn, New York), and Dr. Reynolds reports an interesting case of missed abortion, i.e. the abnormal retention of dead products of conception, in which the Reynolds tokodynamometer was used to record the motility of the uterus. The invention and physiological application of this instrument have been discussed in Year Books Nos. 47, 48, 49, and 50. In the case now described, no spontaneous uterine contractions were observed prior to the use of oxytocic drugs. The details of the successful treatment, being chiefly of clinical interest, need not be summarized here. The Department of Embryology is however gratified when discoveries and devices made within its walls prove useful in practical medicine.

Uterine Contractions after Progesterone

Another application of the tokodynamometer reported by Drs. Rowe-Dutton, Lubin, and Reynolds concerns the effects upon the uterus, in or near the time of labor, of the corpus luteum hormone, progesterone, administered by injection in oil and in aqueous solution, and a watersoluble preparation of the corpus luteum not known to contain the hormone. None of these preparations affected either the frequency of the contractions or the normal dominance of the fundus. The progesterone-containing preparations seemed (as far as can be deduced from the small number of observations) to have a tendency to reduce the general intensity of uterine contractions, and to retard delivery of those women who received them while in labor.

BIOCHEMISTRY AND PHYSIOLOGY OF UTERINE MUSCLE

Three articles published during the year present results of the group investigation of uterine muscle now under way in the Department's laboratories. Two of these papers (Csapo and Corner on contraction of uterine muscle and on shortening of uterine muscle at different temperatures) present conclusions that already have been adequately summarized in Year Book No. 50. A third (Menkes and Csapo) deals with changes in the energy-yielding substances of the rabbit uterus, i.e. adenosine triphosphate and creatine phosphate, during the period of sexual maturation (when the ovarian estrogenic hormone is dominant in action upon the uterus) and after ovulation (when the corpus luteum hormone, progesterone, is dominant). During the course of sexual maturity there is a 73 per cent increase in the high-energy phosphate content of the uterus. After ovulation a further increase occurs. As stated earlier in this report under "Program of Investigations," another step has subsequently been taken by using a new technique which separately measures the metabolic chain of the adenosine tri-, di-, and monophosphates, to discover how their relative concentrations in the uterus are affected by the ovarian hormones.

BIOCHEMISTRY OF DEVELOPING BRAIN AND LIVER

Dr. Louis B. Flexner, while he was a member of the Department of Embryology (1940-1951), led a long series of investigations of the chemical differentiation of the fetal cerebral cortex, intended to show how the development of the brain structure is paralleled by development of chemical substances and of enzyme systems, and other features upon which physiological brain action depends. The results of these studies have been summarized from time to time in these annual reports; several especially valuable group contributions were discussed last year in Year Book No. 50. Another article, fourteenth in the series on "Biochemical and Physiological Differentiation during Morphogenesis," appeared in August 1951, just after Dr. Flexner's departure for his new post. This paper, by Dr. and Mrs. Flexner, reports a study of the concentration of nucleic acid, a substance or group of substances supposed to be intimately concerned in some unknown way with the

synthesis of proteins in living tissues. Dr. Flexner's previous work had prepared him to test this supposition by comparing the concentration of pentose nucleic acid (PNA) with the known development of proteins in brain and liver. In the case of the guinea pig's brain cortex, he and his co-workers have found that a period of intensely active synthesis of proteins begins at the 41st day of gestation, when the processes of the nerve cells develop. In the case of the liver the increase in the proteins has been found to continue during a considerable part of gestation. Comparison of the calculated amount of PNA in the cytoplasm of the cortical nerve cells and the liver cells with the calculated rate of protein synthesis revealed in both organs an initial rise of PNA paralleling an increased rate of protein synthesis, but the increase in PNA does not continue in a proportional relation to the synthesis of proteins. In the guinea pig's brain (and also in fetal rat muscle as reported by other workers), there may even be a decrease in PNA concentration with increased rate of protein synthesis. Flexner and Flexner point out that their attempt to compare the curves of the two substances in the whole cell may be misleading because the synthesis of protein and the presumed participation of nucleic acid in that process may be limited to special regions of the cell. If the Nissl bodies alone are considered, for example, the thesis of a relation between PNA and protein synthesis is satisfied.

DIFFUSION AND APPLICATION OF RESEARCH RESULTS

Conferences. Dr. George W. Corner attended, as delegate of the American Association of Anatomists, a conference on international anatomical nomenclature, held at London, England, May 26–30, 1952. Dr. Corner was elected chairman of the

conference, which was held under the auspices of a subsidiary of UNESCO, the Commission for the Correlation of International Congresses of Medical Science. The eight delegates, representing a major proportion of the anatomists of the world,

were called together to explore the possibility of establishing a single international terminology in place of four systems now in use in various countries. As a result of this meeting, at which a high degree of agreement was evinced, an international revision of the Basel *Nomina Anatomica* of 1895 will be prepared for submission to the next International Congress of Anatomy in 1955.

Dr. Corner took part in a Conference on Population Problems at Williamsburg, Virginia, June 20–22, 1952, held by the National Academy of Sciences at the suggestion of Mr. John D. Rockefeller 3d.

Dr. S. R. M. Reynolds participated in a Conference on Visceral Circulation held at London in July 1951, under the auspices of the Ciba Foundation; and in a Conference on Fetal Welfare, called by the Association for the Aid of Crippled Children, at the New York Academy of Medicine, in June 1952.

Reviews. Dr. Corner's Huxley Lecture, given at Charing Cross Hospital, London, May 28, 1952, which will shortly be published in the British Medical Journal, constituted a review of current knowledge about the ovarian cycle of primates. A significant part of this information was worked out by past and present members of the staff of the Department of Embryology.

Dr. Reynolds prepared for *Physiological Reviews* a summary of our knowledge of the determinants of uterine growth and activity.

In a discourse before the International and Fourth American Congress on Obstetrics and Gynecology, held at New York in June 1950, Dr. Reynolds reviewed present knowledge of the contractility of the human uterus and its physiological basis. He dealt with the architecture of the myometrium (muscular wall of the uterus); the pattern of rhythmic contractility; the

role of the fundus uteri as initiator of the normal pattern; abnormalities of the rhythm and the effects of distention upon the rhythm. Much of this information has been acquired in recent years through investigations by Dr. Reynolds and his coworkers. In a second section he discussed the biochemical studies of Dr. Arpad Csapo and the clues they give with respect to the energy aspects of contraction of the whole uterus.

At the International Jubilee Congress of the French Gynecological Society, Paris, 1951, Dr. Reynolds reviewed the subject of the blood vessels of the ovary in relation to ovarian function, basing his summary largely on research by himself and associates, as reported in Year Books Nos. 47 and 48.

Lectures. In the Johns Hopkins Medical School and Hospital Dr. Corner and Dr. Burns each gave, as usual in recent years, two lectures to the students of anatomy. Dr. Reynolds lectured to medical classes and research groups in internal medicine, obstetrics, pediatrics, and surgery. Dr. Csapo gave three lectures and demonstrations to the third-year class in obstetrics, and three to the gynecological seminar.

Dr. Burns was invited to give the principal address at the rededication of the newly rebuilt biology laboratory of the University of Florida, Gainesville, February 21, 1952. On February 12, he gave a lecture on his recent researches before the combined Departments of Biology and Anatomy of the University of North Carolina, Chapel Hill; and on May 20, he similarly addressed the Department of Anatomy at Yale Medical School.

Dr. Reynolds was invited to address the obstetrical societies of Brooklyn, Los Angeles, and Montreal. He was visiting professor in the Post-Graduate Obstetrical Course at the University of Minnesota, and during the same week gave the Duluth

Clinic Lecture in Minneapolis. He also lectured at the University of Washington Medical School, Seattle, and at Stanford University School of Medicine in San Francisco.

Dr. Csapo was invited to address a group of scientists at Camp Detrick (U. S. Army) on "Actomyosin and Uterine Tension." He spoke also before the Johns Hopkins Biology Club.

Visiting groups. On November 2, 1951 the Department of Embryology was honored by a visit from the Gynecological Visiting Society of Great Britain and Ireland. The party was made up of eleven outstanding specialists, all of them teachers in various hospitals and medical schools of the United Kingdom and Fellows of the Royal Society of Obstetricians and Gynecologists. The staff of the Department arranged a program of demonstrations and experiments.

In December 1951 the Department had the pleasure of entertaining the official party of Directors and Executive Staff of the Carnegie Institution of Washington. In May 1952 the biology classes of several local private schools for girls were given tours of the laboratory with demonstrations of human embryology.

Motion picture film. Following up the great success of the motion picture film on the embryology of the human eye, mentioned in the annual report last year (Year Book No. 50), the American Academy of Ophthalmology and Otolaryngology has undertaken sponsorship of a similar film on the embryology of the human ear. This will be produced by Sturgis and Grant, Inc., of New York City, as was the previous film. The Department of Embryology is already co-operating with the producers and the Academy's committee, by providing technical advice, as well as pictures and models for the use of the animating artists.

Photographic award. A certificate of merit of the Sixteenth Rochester International Salon of Photography was awarded to Mr. Richard D. Grill for a panel of three photographs of a human fetus in utero made in the regular course of work of the Department of Embryology. In one picture of the set infrared photography was very successfully used to reveal the fetus within the unopened membranes.

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DEPARTMENT OF GENETICS

Cold Spring Harbor, Long Island, New York
M. DEMEREC, Director

The past year will stand out in the history of this Department because of the construction of our new laboratory buildings. Although they have not yet been completed, the work is far enough advanced to give a good indication of the modern laboratory facilities we may look forward to utilizing in the very near future. In spite of the restrictions imposed by our present crowded quarters, good progress has been made this year in the research programs of our staff. The most important accomplishments will be briefly summarized here.

RESEARCH

Study of the origin and nature of instability of genic action at a number of different known loci in the maize chromosomes has been continued by McClintock. She has established that, at any one locus of known genic action, different types of instability expression can appear. Each is believed to reflect the operation of a particular chromosomal system, which controls action of the genic components at the locus. The controlling systems appear to be composed of distinct chromosomal units; but, unlike the genes, these units may move from one location to another within the complement. When one such unit moves to a new location, it imposes its specific mode of control of action of the genic components at that location. Ability to recognize the operation of a particular system controlling instability of genic expression depends on its specificity. One of the systems studied is so highly specific that its operation at any locus is readily detected. Experiments designed to introduce this system of mutational control at several selected loci have been successful. The very same system of mutational control has now been obtained at the loci of six different known genetic factors; and comparative studies have been made of its operation at the several loci. At five of these loci, other systems of mutational control have also appeared, so that it has been possible to compare the operation of different systems of mutational control at the same locus. It is now clear that different systems of control of genic expression may arise at any one locus in the chromosome complement, and that the same system may operate at different loci. McClintock has concluded that at least two classes of functional genetic units are carried by the chromosomes: one of them potentially capable of determining a particular course of cellular reactions, the other associated with the control of this potential action.

Hershey and Chase have shown that the first step in the growth of bacteriophage T₂, after the virus has become attached to a cell, is injection of the viral nucleic acid into the cell. This leaves the membrane of the phage particle at the cell surface. The membrane contains most or all of the protein of the phage particle, and has no function in intracellular growth of the virus. They have also found that viral desoxyribonucleic acid contains an unidentified base (probably a pyrimidine) not present in bacterial nucleic acid, and that bacterial nucleic acid contains cytosine, not present in viral nucleic acid. These findings will make it possible to study nucleic acid economy during viral growth in a manner previously thought impossible.

Visconti and Coon have studied genetic recombination in bacteriophage in relation to the growth cycle of virus in the bacterial cell. They find that the proportion of recombinants depends on the time of lysis of the infected cells as well as on the linkage relations between genetic loci. They interpret their results in terms of a drift toward genetic equilibrium in a growing and mating population of intracellular This interpretation of phage "crosses" also explains several other peculiarities of phage genetics. It has been formulated as a quantitative theory by Visconti and Delbrück (in press). Visconti and Coon have also described a physiological change conferring resistance to "lysis from without" upon infected bacteria. This change occurs during the first few minutes after infection. Several methods have been developed to study the kinetics of the process.

Witkin and Fetherston have obtained additional evidence that the cytological nucleus of Escherichia coli may be regarded as a true genetic nucleus, as a result of experiments indicating that the determinants of resistance to bacteriophage are located in this staining body. Studies of the effects of temperature on spontaneous and induced mutations to phage resistance in E. coli were also begun by Witkin and Fetherston. They have shown that spontaneous mutation and cell division have the same temperature coefficient, so that the rate of mutation per generation remains constant over a wide temperature range. The yield of ultraviolet-induced mutations was found to be drastically reduced when the treated cells were grown at 16° C, as compared with the number obtained when the postirradiation divisions were at 37° C. Preliminary results at 25° differed little, if at all, from those at 37°, indicating that the temperature effect may be characterized by a threshold below 25°.

Temperature was found to have no effect on the pattern of delayed appearance of induced mutants, the fraction of the total vield detectable at any given postirradiation division remaining constant over the temperature range investigated. A sensitive period for the temperature effect was also demonstrated. The final yield of induced mutations was shown to be affected by the temperature during only the first postirradiation generation, despite the fact that ten to twelve generations are required to reach the end point. After the first generation, the rate of appearance of induced mutants shows the same temperature response as the process of spontaneous mutation. These experiments provide support for the hypothesis that ultraviolet light induces a metastable condition, leading to a high rate of mutation during the generations following irradiation.

Demerec, Hanson, and Monsees have studied spontaneous and induced mutability in thirty-five different genes of Escherichia coli that determine the organism's requirements for various amino acids. Five of these genes are "mutagen stable"; that is, their rates of mutation cannot be increased by treatment with X-rays, ultraviolet rays, manganous chloride, or β-propiolactone. It has also been determined that manganous chloride and ultraviolet radiation are specific in their mutagenic action, manganous chloride being more effective for certain genes and ultraviolet for others. Delay in the appearance of induced mutants appears to be very common in this material, since it was observed in seventeen out of twenty-one cases tested. The studies of this phenomenon that have so far been completed make it seem probable that a labile condition of the gene, induced by treatment with a mutagen, is responsible for the appearance of mutations during several cell divisions after the treatment. A working hypothesis, developed by Demerec to explain the action of mutagens, postulates that the effect of a mutagen on genes is indirect; in other words, that the mutagen induces changes, in either the cytoplasm or the nucleus, which affect the metabolic processes of treated cells, and that the various members of the genome react differently to the new conditions created by treatment. These conditions persist until the treated cells have passed through one division, which appears to be sufficient to re-establish the original metabolic state.

Additional evidence regarding the differential action of mutagens has been obtained by Galinsky, who observed that small-colony reversions to prototrophy were induced in a glycine-requiring strain of *E. coli* with a higher frequency by ultraviolet radiation than by manganous chloride, whereas the reverse was true of large-colony mutations.

Labrum has made an extensive study of mutability at the streptomycin locus in *E. coli*. He has found that alleles which originate independently differ from one another in some property, such as spontaneous or induced mutability or—in the case of streptomycin-dependent alleles—the amount of residual growth on medium containing no streptomycin. Among seventy alleles investigated, he found no mutagen-stable types, suggesting either that mutagen stability is limited to certain loci or that it occurs more frequently at some than at others.

Demerec and Hanson have made tests of mutagenicity with eight aromatic nitrogen mustard compounds received from Professor A. Haddow, of the Chester Beatty Research Institute in London. Seven were found to be mutagenic, and one, which was not soluble in water, showed no mutagenic effect.

Analysis by Kaufmann has revealed that

shrinkage of chromosomes produced by solutions of pepsin is due to contraction of the partially hydrolyzed nucleoprotein gel, and not to selective action of the enzyme on a matrix protein, as had previously been assumed. Treatment of salivarygland chromosomes of Drosophila with solutions of trypsin has produced microscopically demonstrable evidence of their polytene pattern of organization. Evidence of further subdivision into fibrous subunits has been obtained by Gay in electronmicroscope studies of smears of whole chromosomes. Cytochemical studies by Dr. Nebahat Yakar of cells in which pycnosis had been produced by chemical treatment have shown that the degenerative changes involve dislocation of nucleic acids and proteins. Roots growing from seeds treated with chloranil, which is commonly used as a fungicide, were found to have cells with polyploid numbers of chromosomes and various types of structural aberration.

Studies by McDonald and Moore of the inactivation of dilute solutions of crystalline chymotrypsin by X-radiation have shown that, as with trypsin, the ionic yields for this reaction are not constant, but increase with increasing initial concentrations of the enzyme. They are also a function of the solvent used and of the pH of the solution being irradiated, the pH of optimum stability being approximately 6. These investigators have also shown that dilute solutions of trypsin are inactivated by hydroxyl radicals, the amount of inactivation being related exponentially to the time of exposure. McDonald has found that onion root tips contain an enzyme which depolymerizes desoxyribonucleic acid without the formation of inorganic phosphorus, and which is still present in measurable amounts in roots that have been fixed in Carnoy's fluid, embedded, and sectioned. The rate of depolymerization is

highest between 60° and 70° C, and at pH 5.5 or 6.0, depending on the solvent in which the reaction takes place.

Paigen and B. N. Kaufmann, working on the effects of exposure to X-rays, have studied the nucleic acids of mouse liver during the postirradiation period. They have concluded that the net effect of radiation on the quantities of pentose nucleic acid and desoxypentose nucleic acid is either an increase or a decrease of both nucleic acids, depending on the time after irradiation at which measurements are made, and that the radiation does not affect the interconversion of nucleic acids and nucleotides. Determinations of the bases present in the two nucleic acids after exposure to X-rays have revealed no qualitative change in the nature of the nucleic acids during the postirradiation period. Paigen, continuing his studies of mitochondria, has found conclusive proof of the existence of three types, and has accumulated information about the chemical nature and enzymatic properties of these

Again this year the research program of the Department was expanded with the help of three grants. Two of these came from the U. S. Public Health Service, one in support of research by Kaufmann and McDonald, and the other in support of Hershey's work. A third grant was received from the American Cancer Society, upon recommendation of the Committee on Growth of the National Research Council, in aid of research by Demerec and Witkin.

RETIREMENT

After thirty-eight years' service as a staff member in the Department, Dr. E. Carleton MacDowell is retiring in September 1952, having reached the obligatory retirement age for members of the Institution.

MacDowell was attracted to Cold Spring Harbor while he was still a zoology student at Swarthmore and Harvard, and he spent the summers of 1908 to 1910 taking courses at the Biological Laboratory and doing research under C. B. Davenport, who was then Director of both the laboratories. During that period he completed a study of the embryology of syndactyl chicks. As a graduate student at Harvard he worked under Professor W. E. Castle. one of the pioneer geneticists, whose experimental study of heredity in small mammals was supported by the Carnegie Institution. MacDowell's thesis, Size Inheritance in Rabbits, was published in 1914 as Carnegie Institution of Washington Publication 196.

After spending two years as an instructor in zoology, one at Dartmouth and one at Yale, MacDowell settled in Cold Spring Harbor, in 1914, as a staff member of this Department. Although after leaving Harvard he had carried out some work with Drosophila, studying the effect of selection on number of bristles, he now turned back to research with mammals, which he has continued for almost forty years. His first problem at the Department was a study of the effects of grandparental alcoholism on learning capacity and reproduction in rats. Some influence on learning capacity was observed; and it was found that alcohol, by selecting against the poorer breeders, improved the reproductive capacity of the surviving descendants.

During this early period at Cold Spring Harbor, from 1914 to 1928, MacDowell began his work with mice, investigating the effects of alcohol on reproduction and sex ratio. By means of a surgical technique that he developed for determining the number of ova at the start of a given litter, he was able to ascertain the total prenatal mortality, and to discover that litters with

no loss showed a primary sex ratio of one male to one female.

These investigations led MacDowell to become interested in studies of growth. With Charlotte MacDowell, he made a very exact determination of the growth curve of mouse embryos. Later, with Dr. Ezra Allen, he worked out growth curves for suckling mice; and with Professor P. E. Smith he studied the growth of mice belonging to a dwarf strain. MacDowell and Smith discovered that this dwarfism was caused by inherited pituitary deficiency; and their study demonstrated the primacy of the pituitary as related to the development and functioning of the other endocrine glands.

A turning point in MacDowell's research came in 1928, when he found that about 90 per cent of the mice of his strain C58, which had already been inbred for 18 generations, developed leukemia, whereas in the other strains of his colony leukemic individuals occurred but rarely. The unusual opportunities this material offered for pioneer investigation in a new field were fully realized by the administration of the Institution, and MacDowell was given greatly increased facilities for research. He was very successful in establishing collaborative relations with pathologists and physiologists, particularly with Drs. M. N. Richter, J. Victor, and Max Bovarnick, of the College of Physicians and Surgeons of Columbia University; and, together with Dr. J. S. Potter and Miss M. J. Taylor, his associates at Cold Spring Harbor, he carried on an intensive research program dealing with basic problems of leukemia.

This research established that leukemia is a malignant growth parallel to cancer, and that, like cancer, it can be transmitted from one individual to another by transplantation of affected cells. The studies

revealed intrinsic differences among transplants from different spontaneous cases, and intrinsic changes during a long series of subtransplantations. In the course of this work many lines of leukemia, characterized by specific differences, were isolated. It was found that, whereas mice of the C58 strain were susceptible to transplanted leukemia, mice from other inbred strains were not. In the case of line I, crossing experiments showed simple Mendelian segregation of resistance and susceptibility, indicating that one important recessive gene is responsible for the resistance. That the ratios obtained actually represented genetic segregation was proved by breeding tests in which a mouse was classified by the proportion of resisters among its offspring instead of by the results of its own inoculation. A similar analysis of resistance to spontaneous leukemia gave evidence of genetic segregation involving several genes.

A new approach to the study of the extremely complex problem of leukemia was made possible by the discovery that a maternal factor for resistance to spontaneous leukemia may be transmitted to mice of a susceptible strain by the milk of old nurses belonging to the resistant strain StoLi. It was also found that the resistance factor may be transmitted *in utero* when old StoLi mothers are bred by susceptible-strain males.

Methods have been developed for inducing resistance to certain transplanted leukemias. Resistance to transplanted line-I_b leukemia, in terms of the current interpretation, depends upon an "X-factor," which is separable from line-I_b cells by sonic vibration.

Although MacDowell retires this year, plans have been made for a continuation of his studies of resistance to transplanted leukemia, in collaboration with Dr. J. Victor, who is now at the Chemical Corps

Biological Laboratories in Frederick, Maryland. Studies on the chemical nature of the maternal resistance factor will be pursued in collaboration with Dr. Samuel Graff, at the College of Physicians and Surgeons, Columbia University.

As a member of the staff of the Department of Genetics, MacDowell has taken part in several activities of the Institution. He was particularly active in planning the exhibits formerly held in Washington in conjunction with the annual meeting of the Board of Trustees. For a number of vears he was a member of the Committee on Exhibits, and also its co-chairman and chairman. For one year he held the chairmanship of the lecture committee. At the request of the administration, he visited the Chichen Itza project in Mexico, as well as the Institution's western laboratories, and attended a conference on Institution affairs held in Tucson in 1929.

In 1931, shortly after the research on leukemia began, Miss Martha J. Taylor came to work with MacDowell as research assistant. During the more than twenty years she has remained at the Department, Miss Taylor has taken a responsible part in many of the experiments with leukemia. In addition, she has made valuable contributions in various aspects of our community life, participating in the management and redecoration of the dormitory and for two years taking charge of the dining room. At the termination of MacDowell's connection with the Department, she will leave for a position with the Chemical Corps Biological Laboratories in Frederick, Maryland.

Fellows

This year our Department had two Carnegie Institution Fellows, Dr. Kenneth Paigen, continuing his fellowship for a second year, and Dr. N. Visconti, of the

Institute of Chemistry of the Polytechnical School, Milan, Italy, who was with us last year as a guest. Paigen, on the termination of his fellowship in September 1952, will take a research position at the Peter Bent Brigham Hospital and Harvard Medical School. Visconti has been appointed a staff member of the Department, and will continue his research on the genetics and biology of bacterial viruses. Until March 1952, Dr. Nebahat Yakar, of Istanbul University, worked on cytogenetic problems with Kaufmann, as a Fellow of the Turkish Government.

CO-OPERATIVE WORK

Close co-operation with the members of the Biological Laboratory has had a stimulating effect on research at the Department. The study being conducted by B. Wallace and J. C. King, dealing with the effect of continuous exposure to ionizing radiation on the genetic constitution of Drosophila populations, has now progressed through more than eighty generations. The results indicate that radiation has no detectable detrimental influence on the viability of populations. In June 1952, King began a study of the mechanism responsible for the origin of insect resistance to insecticides. In the first stages of this study, Drosophila is being used as experimental material.

V. Bryson's studies of spontaneous and induced mutability in *Escherichia coli* have contributed further evidence that mutants which involve the same character but originate independently are not alike. A comparison by Bryson of spontaneous and ultraviolet-induced mutations to phage resistance has revealed both quantitative and qualitative differences. E. Rosenblum has studied the physiology of *E. coli* treated with certain mutagens. He has found that treatment with manganous chloride retards the rate of oxidation of

glucose, whereas ultraviolet irradiation does not affect it.

W. Szybalski, continuing the study of bacterial resistance to antibiotics, has extended his investigations to include the isoniazid compounds, which were recently found to be effective against the tubercle bacillus. He has found that high resistance to isoniazids develops quickly, in one step, in which respect these compounds resemble streptomycin.

Members of our staff have co-operated in various ways with scientists at other laboratories. We have attended seminars and meetings at Brookhaven National Laboratory and Columbia University. Dowell has utilized sonic equipment at Brookhaven to break open leukemic cells, and Demerec has used that Laboratory's high-capacity X-ray equipment for irradiating bacteria. Kaufmann has co-operated with Dr. T. F. Anderson, of the Johnson Foundation of the University of Pennsylvania, in electron microscope studies. Visconti spent about a month with Dr. M. Delbrück at the California Institute of Technology in Pasadena, completing a mathematical analysis of genetical recombination in bacteriophage begun the previous summer while Delbrück was staying at the Biological Laboratory.

MEETINGS AND LECTURES

The Seventeenth Cold Spring Harbor Symposium, held early in June, brought together more than 120 scientists interested in the physiology of the neuron. A considerable number of the participants came from abroad.

Hershey spent part of the 1952 summer season in Europe. He participated in a Symposium on Immunochemistry at the meeting of the Society of Biological Chemists in Paris, and in a conference on bacteriophage sponsored by UNESCO. Demerec spent about four weeks in England and Scotland, visiting laboratories in Glasgow, Edinburgh, Cambridge, London, and Bayfordbury, and at Oxford, where he gave an invitation paper at the Symposium on Evolution organized by the Society for Experimental Biology and the Genetical Society of Great Britain.

At various times throughout the year members of the staff were invited to discuss their research in seminar meetings at other institutions. Demerec spoke at the University of Pennsylvania, the University of Glasgow, Hammersmith Hospital in London, and the John Innes Horticultural Institution in Bayfordbury; Hershey at Princeton University; McClintock at Yale University, the University of Illinois, and Columbia University; McDonald at Union College and the General Electric Laboratories in Schenectady; MacDowell at the National Cancer Institute in Bethesda; and Visconti at the California Institute of Technology.

OTHER ACTIVITIES

In April 1952, Kaufmann was elected to membership in the National Academy of Sciences, and Demerec to membership in the American Philosophical Society.

For the eighteenth year Demerec compiled and edited the *Drosophila Information Service*, a mimeographed bulletin issued by the Department and containing material, contributed by *Drosophila* research workers, which is of interest to a large group and is not likely to be published elsewhere. The directory section of the November 1951 issue listed 119 laboratories and 544 research workers co-operating in the project. A similar circular for research workers in genetics of microorganisms, the *Microbial Genetics Bulletin*,

was compiled and edited for the third year by Witkin.

Our Drosophila Stock Center, under the care of Mrs. G. C. Smith, sent out a total of 1108 cultures to research and teaching laboratories. Of these, 106 shipments were

to Europe, three to South America, and one to Japan.

The Department's library, with Mrs. H. H. Wheeler in charge, acquired 206 books during the year and received 274 periodicals and serial publications.

MOUSE LEUKEMIA

E. C. MacDowell, M. J. Taylor, and B. Stuard

In the early years of the experimental study of malignant growths, an enormous amount of research was devoted to tumor resistance. That a transplanted tumor could induce resistance to itself seemed to offer great hope for a practical application. But as it became clear that the capacity to induce resistance was neither general nor characteristic of certain types of tumor, but applied only to certain specific transplanted tumors, interest faded, and many investigations were terminated before reaching interpretable results. Many phenomena of biological interest had, however, been observed. In this laboratory many similar phenomena have appeared in the transplanted leukemias derived from spontaneous cases in strain C58, and their study has been reported in these Year Books over a period of more than twenty years. The fact that one highly inbred strain of mice has provided both the lymphatic leukemia and the hosts for transplantations eliminates questions such as those that arise concerning the comparability of previous results obtained with a wide variety of malignant growths, host strains, and laboratory conditions and methods.

An outstanding result of this work, which is also its major challenge, is that resistance may be induced to some of these lines of transplanted lymphatic leukemia and not to others. The leukemias that are not resistable include first transfers from spontaneous cases, as well as the spontaneous cases themselves, whose incidence

is not reduced in hosts immunized to a certain transplanted leukemia, no matter how firm the immunization. Indeed, even after a spontaneous case in such an immunized host has become well developed, a lethal dose of the transplanted cells will be destroyed, while the cells of spontaneous origin continue to multiply.

Discovery of the nature of the mechanism responsible for resistance to those leukemias that are resisted would offer a clue to the reason other leukemias are not resisted, and at the same time might illuminate general processes of resistance as well as the nature of malignant growths.

The capacity of line I leukemia to induce resistance to itself has been known since 1934. Interest has long been centered on methods of bringing this capacity into action, and on the mechanism involved in its operation. Thanks to the long-unsuspected co-operation of a contaminating virus, a sufficiently diluted dose of line-I cells would induce a certain amount of resistance, which permitted the mouse to survive progressively larger doses. After the virus had been discovered and eliminated (line Ib), resistance to a moderate test dose of these leukemic cells could still be established in three days by one dose of finely minced normal tissue (embryonic or adult) from a foreign strain (StoLi). Resistance also followed one dose of line-Ib cells that had been heated for 7-14 minutes in a 46° C bath. And, finally, resistance followed one dose of tissue from

a mouse of the strain of origin (C₅8) after it had been firmly immunized by massive doses of line-I_b cells. These procedures seemed quite diverse, and appeared to utilize different mechanisms in the first step of inducing resistance. But the resistance following survival of the first test dose of living line-I cells seemed to be the same phenomenon in every case, and clearly a form of active resistance.

Since the development of active resistance is generally supposed to require two or three weeks, some other mechanism (or possibly several other mechanisms) seemed required to account for resistance to the initial dose of living line-Ib cells. But the work of the past year leads to the interpretation that, even in the process of surviving the first dose of living cells, active resistance plays the major role. The evidence indicates that the unquestionably active resistance following the first test dose is developed rapidly, and that some factor from the cells themselves stimulates a defensive reaction in the host even in the process of resisting the first dose of line-Ib cells.

One standard dose of line I_b (10⁸ ± cells) heated 14 minutes in a 46° C bath will enable a mouse to resist a moderate dose (about 20,000) of unheated line- I_b cells, but not a dose 256 times larger. When this larger dose was given 2 days after the smaller one, however, a third of the mice survived (5/15); and when it was given 4 days after, all survived (15/15).

That the cells inactivated by heat (14 minutes at 46° C) induce a host reaction is suggested by the increase in resistance during the 2 days after treatment. The proportion of survivors is 23 per cent, 65 per cent, and 99 per cent, respectively, when test doses of 20,000 cells are given at once, after 1 day, and after 2 days.

If the X-factor responsible for this reaction is not a product of the heated leukemic

cells themselves, it is very closely associated with them, for washing them as many as four times—each time discarding the supernatant after centrifugation, and resuspending the cells in saline—did not modify the results. This was true not only for 14-minute but also for 7-minute heating; and the dilution effect (Year Book No. 50, 1950–1951), which had been the first direct indication of an X-factor in some way separate from living cells, was still demonstrable.

The fact that almost all the red cells in the heated material could be eliminated by differential centrifugation without reducing the resistance emphasized still further the close association of the X-factor with leukemic cells. And, finally, similar results were obtained with unheated cells that had been ruptured by sonic vibration, as described below.

The cordial co-operation of the Biology Division of the Brookhaven National Laboratory made it possible to perform several experiments with unheated, washed line-Ib cells after subjecting them to sonic vibration. Under the conditions used, all the cells were not disrupted, as was determined both microscopically and by the production of leukemia; but after the cells and the heavier debris had been almost completely eliminated by centrifuging for 15 minutes at 600 × gravity, the following results were obtained. A test dose of 20,000 untreated line-I_b cells was resisted (57/60) when given about 2 days after the supernatant from the vibrated material, but when it was given at the same time there were no survivors (0/10). A test dose of 1000 cells, given at the same time as the supernatant of the vibrated material, was resisted (7/9). None of the controls survived the test doses alone.

Although the similarity of these results to those obtained with heated cells does not finally demonstrate that the same X- factor operates in both situations, this is the simplest interpretation. If the same factor does operate, the implication is that the heat, rather than inducing the production of a new or modified substance, merely inactivates cells and thereby permits the escape, after inoculation, of a factor already present, whereas sonic vibration frees this factor before inoculation. The X-factor thus does not depend on integration with cellular organization for its effective delivery to a host, and therefore it is available for identification.

If resistance is called active when it is induced by something provided by the pathogen, the initial resistance in this case is active by definition. One set of results, however, may seem to cast doubt on this interpretation. Active resistance, as usually known, persists; and after even a few living line-Ib cells have been survived, the resistance does persist for three weeks or more, whether these living cells are given as a test dose or are cells that have survived 7-minute heating or the 1000 unheated cells that are mixed with those heated for 14 minutes. But without the addition of some unheated cells, the host reaction induced by one dose of the cells inactivated by 14 minutes at 46° C, after reaching a maximum in 2 to 3 days, fades out very quickly and is nearly gone in a week. Although this rapid disappearance may suggest some form of passive, rather than active, resistance, it is clear that this case is peculiar because of the small amount of the X-factor available in the single dose of 14-minute-heated cells, in comparison with the amount finally contributed by the inactivation of living cells after they have multiplied extensively. For it was shown years ago in this laboratory that, when first injected, the cells of a test dose increase to the point of forming widespread lesions, no matter how firm the immunization of the host and how certain its survival. In

the end, the host reaction destroys all the leukemic cells, so that an excess of X-factor is supposedly released and a reserve of the host-produced anti-line I substance remains.

A picture thus emerges of the phenomenon of induced resistance to line-I leukemia not as an absolute or static state capable of destroying invading cells as they enter, but rather as a complex, kinetic process, in which the growth of the invading cells themselves plays a critical part—a process that is repeated at every challenge. In more explicit terms, the leukemic cells of line I_b appear to carry an X-factor, which, when freed, stimulates the host to vield an anti-line I substance. This substance inactivates some leukemic cells, which thereupon liberate more X-factor, while other cells continue to multiply. A chain reaction is started, but whether it moves fast enough to save the life of the mouse depends on various conditions. If these conditions provide a sufficient head start for the host reaction in relation to the size of the dose of living cells, the mouse will survive. In most cases there is enough X-factor in one standard dose of cells heated 14 minutes at 46° C to give the host reaction the advantage needed to restrain and finally eliminate a leukemic population that started with 1000 cells, even if these are inoculated on the same day as the heated cells. But if 20,000 cells are inoculated at once, the mouse dies in most cases, although the total amount of antileukemic substance formed by the host may be as great as or even greater than before. If the host reaction is given a head start of 2 days, the leukemic population produced by the inoculation of 20,000 cells will eventually be eradicated. If the conditions do not give a clear advantage to the host reaction or to the leukemic cells, the balance between death and survival may be decided in individual cases by subtle

variables, which can be controlled only statistically. Sometimes the closeness of this contest is indicated by delay of death beyond that of the controls, and sometimes by recovery after the spleen has become palpable and even after it has reached a size found in leukemic autopsies.

The studies on experimental leukemia that have been regularly reported in this Year Book have made something of a record for persistence and continuity, unbroken by the periods of depression and war. The mice have been bred with the same care and diet and procedures since 1924. The leukemic strain (C58) has been carried through 96 inbred generations, and line-I leukemia through more than 2000 successive hosts; 12,000 transplantation experiments have been performed. Although this report terminates the series, there is no end to the vistas leading to more specialized fields, and there are prospects

that work in some of these directions will be pursued more extensively and with more specialized techniques. For the past two years Dr. Jerome T. Syverton, of the Department of Bacteriology and Immunology of the Medical School of the University of Minnesota, has been verifying and pursuing certain of these findings on the nature of induced resistance, employing our materials and drawing on our experience. Another of these leads—the maternal resistance factor (Year Book No. 50 and earlier)—has been chosen by Dr. Samuel Graff, of the College of Physicians and Surgeons, Columbia University, for a major chemical project. This undertaking seems highly appropriate, for from the College of Physicians and Surgeons came the first diagnosis of leukemia in strain C58, as well as the proposal for the cooperative investigation, with Dr. Maurice N. Richter, that initiated these studies.

BACTERIAL GENETICS

M. Demerec, E. M. Witkin, E. L. Labrum, I. Galinsky, J. Flint Hanson, H. Monsees, and T. H. Fetherston

During the past year the interest of our group was again focused on studies of spontaneous and induced mutability in B strains of Escherichia coli. We were able to demonstrate specificity in the action of certain mutagens. We made further investigations of "mutagen stability," that is, the failure of some loci to respond to the action of mutagenic agents. We obtained additional evidence that the determinants of phage resistance are located in the nuclear body. We studied the effect of temperature on spontaneous and induced mutability, and found that the spontaneous rate was not affected, over a wide temperature range, but that the yield of ultravioletinduced mutants was very much smaller when treatment took place at 16° C than when it was given at 37° C, and, more-

over, that a similar result was produced when the low temperature was applied after irradiation but before the treated cells had divided. We obtained evidence that the pattern of delay in the appearance of induced mutants is specific for the locus, and is due to an induced change in the gene from a stable to a labile state. We studied various aspects of mutability at the locus responsible for streptomycin resistance. Our studies of induced mutability have now progressed to the point where we can postulate a sequence of events occurring in cells treated with a mutagen. Finally, we conducted tests to determine the degree of mutagenicity of several compounds used in cancer research.

In addition to the workers named above, our group included Mrs. G. C. Smith, who

had charge of the *Drosophila* colony, and Miss Jeanne Hyne, Miss Margaret Baker, and Mrs. Katherine M. Main, who washed and sterilized the glassware used in work with bacteria and bacterial viruses. During the summer, Mr. Thane Asch assisted in research with bacteria.

Our work received partial support from a grant-in-aid from the American Cancer Society upon recommendation of the Committee on Growth of the National Research Council.

REACTION OF GENES TO CERTAIN MUTAGENS

Last year (see Year Book No. 50, 1950-1951, pp. 184-185) we discovered the phenomenon that has since been called "mutagen stability," when we found that in two histidineless strains of E. coli the rate of reversion to nondeficiency could not be increased above the spontaneous level by treatment with any of three potent mutagens, manganous chloride, ultraviolet radiation, and β-propiolactone. In order to find out whether mutagen stability was limited to histidine deficiencies or was a more general phenomenon, Demerec, Hanson, and Monsees undertook an investigation involving 33 auxotrophic strains (mutant strains requiring growth factors), in which studies were made of mutations ("reversions") to the prototrophic or nondeficient form, both as they occurred spontaneously and as they were induced by manganous chloride, ultraviolet radiation, or β-propiolactone. These 33 strains represented 35 separate mutational occurrences of nutritional deficiency, each resulting in a certain growth-factor requirement-or, in one case, a double requirement. The growth factors involved were eleven amino acids.

Our experimental results clearly showed considerable differences among the various genes included in our study, with regard to rates of spontaneous and induced mutability. They suggested that spontaneousand induced-mutation rates are both determined by the allelic form of the gene present at a certain locus.

We found 3 more mutagen-stable auxotrophs among these strains—one tryptophaneless, one methionineless-threonineless, and one prolineless—making 5 cases all together. Since 5 out of 35 deficiencies tested were mutagen stable, it can be inferred that mutagen stability is a fairly widespread phenomenon; and, since it was found in auxotrophs requiring four different amino acids, it is evidently not limited to any one locus.

Work with these auxotrophs also served to extend the observations on another striking feature of induced mutability, namely, the phenomenon known as delayed appearance of induced mutants. Tests were completed with 21 of our amino acid deficiencies, and revealed this "delayed effect" in all but 4 cases. On the basis of what is known at present, the most plausible explanation of the delayed effect seems to be that mutagenic agents induce an instability in the genome of treated cells, which persists throughout several divisions; and that each locus of the genome has its own characteristic pattern of expression, which is independent of the mutagen initiating the reaction.

A comparative study was made of the efficacy of similar treatments with manganous chloride, ultraviolet radiation, and β -propiolactone in inducing reverse mutations of our series of 35 amino acid deficiencies. This showed that in most cases β -propiolactone was least potent in its mutagenic action, although in one instance it induced more mutations than either of the other two mutagens. Of these two, manganous chloride proved to be the more potent in some cases, ultraviolet in others.

The data shown in table 1 illustrate these points. We can infer that the variations in relative mutagenicity are not due to differences in cell physiology or permeability among the different strains, from the findings with strains, such as 12-72, that carry more than one auxotrophic mutation. If

TABLE 1

Frequencies of reversion to nondeficiency induced by similar treatments with manganous chloride (MnCl₂), ultraviolet (UV), and β -propiolactone (β -pl) in several amino acid—requiring strains of Escherichia coli

STRAIN		INDUCED R	×10 ⁻⁸	
STRAIN	DEFICIENCY	MnCl ₂	UV	β-pl
12-33	Methionine	940	1,020	8
WP-8	Lysine	2,390	2,190	232
Sd-4-55	Tryptophane	10,200	1,800	60
12-36	Threonine	2,185	435	55
M-4	Tryptophane	448	10,700	217
D-84	Arginine	63	4,600	16
12-11	Methionine	1,333	1,020	8,130
12-72	Leucine	594	57	28
	Phenylalanine	11	100	3

physiological conditions of the cells were responsible for the observed differences between separate strains, we could expect two deficiencies occurring in the same strain to react in the same fashion to the same mutagen. Ample evidence is available to show that this is not the case. In strain 12-72 (table 1), where it was possible to observe independent reversions from deficiencies for leucine and phenylalanine, it is evident that each of the two loci involved reacted to the mutagens in its own specific manner. Treatment with manganous chloride and with β-propiolactone induced more leucine than phenylalanine reversions, whereas ultraviolet treatment produced the opposite result. Thus our data support the assumption that the action of a mutagen on genes is specific; in other words, that treatment with a particular mutagen produces a certain change in the cytoplasm or nucleus (or both) which affects different genes of the genome differently.

With the information now available we are in a position to formulate a working hypothesis regarding the sequence of events occurring in cells treated by a mutagen. All the evidence indicates that mutagens do not produce a direct effect on genes, but that their action is indirect, probably through some change in the cell metabolism. The best support for such an assumption is supplied by the observations (1) that the effects of consecutive treatments with two mutagens, ultraviolet and manganous chloride, are not additive (Year Book No. 50, pp. 182-183), and (2) that the effect of a mutagen can be modified by certain other treatments applied subsequently. We have observed that, whereas treatment of streptomycin-dependent (Sd-4) bacteria with manganous chloride at 1° C normally results in a low frequency of reversion to nondependence (172×10⁻⁸ in one experiment), bacteria from the same treated sample kept at 37° C in a 0.015 M solution of NaCl mutated with a considerably higher frequency (6330×10^{-8}) . In another set of experiments, when treated bacteria were washed in either water, 0.15 M NaCl, or 0.15 M MgCl₂, the induced-mutation frequency was decreased from 5000 per 108 to 2420, 2060, or 1890 per 108, respectively.

This modification of the action of a chemical mutagen by subsequent treatment of the exposed cells might be attributed either to an effect on the primary action of the mutagen—assuming that sufficient quantities of manganous chloride remained in the cells after washing, and that posttreatment influenced its activity—or to an effect on the mutagen's secondary

action, that is, on metabolic processes affected by it. The first possibility is eliminated, however, by results of experiments with ultraviolet radiation. Since the primary mutagenic action of radiations ends with the end of the exposure period, posttreatment could be effective only through an effect on secondary action. And Witkin reports (this Year Book) that when ultraviolet-irradiated bacteria are exposed to low temperature after treatment, the mutagenic effect of the ultraviolet is considerably less than when they are kept at a high temperature. Another very interesting feature of Witkin's study is the finding that the effect of the ultraviolet treatment can be modified only up to about the time the irradiated cells undergo first division.

Thus it appears that treatment with a mutagen induces some change in the treated cells, which in turn affects the genome. Studies with manganous chloride, reported last year, have suggested that the chemical is adsorbed to certain sites in the cells, which may be furnished by either nucleic acid or proteins. Different members of the genome react differently to conditions in cells created by the treatment. Some may be completely unaffected (mutagen stable), while others may be affected to various degrees. Apparently each member of the genome exhibits a specific type of reaction toward mutagencreated cell conditions. So far, we have been able to detect such specificity of reaction only by observing the different frequencies with which various genes mutate in mutagen-exposed cells; but there is no reason to assume that other types of difference, perhaps qualitative in nature, do not exist. The indications are that conditions produced by a mutagen may persist in treated cells until they divide, and in the meantime may be modified by application of certain other agents.

In the environment produced in cells by mutagenic treatment, most members of a genome mutate at a rate higher than normal. Our results suggest that a mutation is initiated by transformation of a gene from a stable to an unstable state. It appears, moreover, that the duration of the unstable state, in terms of number of cell divisions, differs for different genes and is locus specific. Our results indicate that the conditions created in cells by mutagens are capable of inducing the unstable state, but that once this is accomplished, the pattern of instability is gene-determined and does not reflect differences in the mutagenic agents employed or the concentrations or doses used in treatment.

DIFFERENTIAL REACTION OF LOCI TO MUTAGENS

A nutritionally deficient strain of E. coli requiring serine or glycine and known as WP-9 has been investigated by Galinsky. This strain yields two types of reversion mutant, both spontaneously and as a result of mutagenic treatment. These mutants have been termed "large-colony" "small-colony," in accordance with the appearance of the mutant clones when grown on minimal-agar plates, and both represent reversions to prototrophy. The spontaneous-mutation rate is low in both cases, being about 0.32×10^{-8} for the large-colony type and 0.062 × 10⁻⁸ for the small-colony type. It appears probable that two loci are involved, since the patterns of delayed appearance of induced mutants differ markedly in the two cases. Only a few generations are required for the appearance of all induced large-colony mutants, but the entire yield of induced small-colony mutants has not yet been manifested by the eleventh generation—beyond which it is impractical to go because of technical difficulties. The fact that both mutations

can be studied in the same strain tends to eliminate some of the genetic and physiological variables encountered in comparisons between different strains.

Sensitivity to the action of mutagenic agents differs in the two cases, as evidenced by the consistently higher number of smallcolony mutants induced even though the spontaneous-mutation rate is lower for this type than for the large-colony type. The small-colony mutants were induced by manganous chloride with an average frequency of about 6000 per 108 treated cells, and by ultraviolet radiation and β-propiolactone with an average frequency of about 2000 per 108. The large-colony mutants were induced with the following frequencies: manganous chloride, about 650 per 108 treated bacteria; β-propiolactone, about 140 per 108; and ultraviolet, only about 2 per 108.

This supports the conclusion, previously reached by Demerec on the basis of work with other strains of *E. coli*, that various mutants respond differently to different mutagenic agents.

MUTABILITY IN STREPTOMYCIN-DEPENDENT STRAINS

The recent studies of mutability by Demerec, Hanson, and Monsees have dealt with reversions in a group of specific amino acid-deficient mutants of E. coli. The mutations observed in a number of these strains presumably involved changes at specific loci. In order to gain more information on mutability, Labrum investigated the genetic behavior of a group of streptomycin-dependent (Sd) mutant strains of E. coli. The mutations observed were reversions from streptomycin dependence to nondependence. Previous crossing experiments with the K-12 strain of E. coli had indicated that mutations involving streptomycin resistance and dependence occur at a single locus. Therefore the various Sd strains of independent mutational origin used in this work presumably represented different alleles at a single locus. The purpose of the investigation was to compare the mutation rates and patterns of delayed appearance of induced mutants exhibited by different Sd strains, and to discover whether or not the phenomenon of mutagen stability could be detected among these strains.

Manganous chloride was the mutagen employed throughout the investigation. The frequencies of spontaneous and induced reversion were determined for 70 different Sd strains. These data showed that none of the 70 strains was mutagen stable; in other words, induced mutations were detected in every strain that produced spontaneous mutations. From this it was concluded that if mutagen-stable types do exist among Sd mutants, they are certainly rarer than among the amino acid-deficient strains.

These results indicated further that practically no two strains were alike with respect to their spontaneous- and induced-reversion frequencies. Spontaneous frequencies among the 70 strains ranged from zero to 16,800 per 10⁸ cells plated, and the induced frequencies from zero to 206,000 per 10⁸ cells plated. These differences in reversion frequencies may have been a reflection of differences in mutation rates among the various strains; but other factors were also involved.

One factor was the number of residual cell divisions that the Sd strains were able to undergo on plain agar lacking streptomycin. For example, in several strains tested, no reversions were detected when either treated or untreated samples were plated on plain agar. Microscopic observation of the plates revealed that the cells had failed to divide, thus preventing the

expression of any spontaneous or induced mutations. On the other hand, certain strains that showed high reversion frequencies were able to pass through 6 or 7 residual divisions on plain agar.

A second factor that could cause differences in the observed MnCl₂-induced reversion frequencies was variation in the proportion of cells surviving treatment in different experiments. Survival varied from 10 per cent to 80 per cent, even though care was taken to perform all experiments in the same manner. Past experience with the use of manganous chloride in this laboratory has indicated that some correlation exists between survival and the frequency of induced mutations.

Further experiments were performed to determine whether or not 15 different Sd strains differed significantly with respect to their mutation rates. Each strain was tested on three different days. The results showed a high degree of variability among the mutation rates for each strain determined at different times. It was evident, however, that significant differences did exist. Certain strains, such as Sd-U3, consistently showed relatively low mutation rates $(2 \times 10^{-8} \text{ for spontaneous and } 8 \times 10^{-5}$ for induced); whereas Sd-S12 exhibited extremely high rates (about 140×10-8 for spontaneous and 240×10⁻⁵ for induced). Most of the other strains had rates intermediate between these two extremes.

An analysis was undertaken of the patterns of delayed appearance of induced mutants in the different strains. Twelve strains which pass through a minimal number of residual divisions were selected. For technical reasons only three of these proved to be well suited to the experimental procedures used. The results indicated that in each of these strains all induced mutants had appeared by about the fifth or sixth division. Fifty per cent

had appeared after 3 to 4½ divisions. It was therefore concluded that these three Sd strains exhibited similar patterns of delayed appearance of induced mutants.

An attempt was made to determine the pattern of delayed appearance in the case of induced mutations from streptomycin sensitivity to resistance and dependence. No conclusive results were obtained, however, since the experimental procedures employed proved to be too insensitive for this system.

Nuclear Segregation and the "Delayed Effect"

Further evidence was obtained by Witkin and Fetherston concerning the genetic functions of the bacterial nucleus. It had previously been demonstrated by Witkin and Schwartz (Year Book No. 50) that the genetic factors controlling the fermentation of lactose are located in the body designated as the nucleus of the bacterial cell. The results to be described below support the hypothesis that the determinants of resistance to bacteriophage also reside in the nuclear element. These experiments show in addition that nuclear segregation plays an insignificant role in the delayed appearance of induced phageresistant mutants.

Populations of strain B/r of *E. coli* differing in the average number of nuclei per cell were obtained simply by selecting cultures of different ages. "Resting" (24-hour) cultures, grown with aeration in a simple synthetic medium, consist primarily of uninucleate cells, as determined by microscopic observation of properly stained samples. Cultures in the late lag phase contain very few uninucleate or binucleate cells, most of the cells having four or eight distinctly separate staining bodies. These two types of culture were used in the experiments, and will be referred to as "uni-

nucleate" and "multinucleate" populations.

Saline suspensions of uninucleate and multinucleate populations having a titer of about 108 cells per milliliter were irradiated with ultraviolet light, using a dose of 800 ergs per square millimeter. Samples of the irradiated suspensions were spread on nutrient-agar plates previously warmed to 37° C, and the plates were incubated at 37°. After various intervals of time, sets of six plates were withdrawn from the incubator and chilled rapidly to arrest growth. Two of the six plates were washed with 10 ml of saline and the wash fluid was assayed, to determine the total number of bacteria on the plates; the four remaining plates were sprayed with an aerosol of a suspension of bacteriophage T₁ having a titer of at least 10¹⁰ particles per milliliter, to ascertain the number of phenotypically demonstrable clones of bacteriophage-resistant mutants. The time intervals for withdrawal of plates from the incubator for washing and spraying were based upon previously established growth curves of the irradiated populations on the plates, and were designed to cover the range from o to 13 generations. The data obtained from numerous experiments of this type permitted the construction of "expression curves" (fig. 1) for uninucleate and binucleate populations, in which the number of T1-resistant clones per 108 irradiated cells was plotted against the number of postirradiation generations. These curves are a measure of the pattern and extent of delayed appearance of the induced mutants—the final yield of induced mutations and the number of generations required to achieve its complete manifestation being easily discerned by the leveling-off point. The curves shown here have been corrected for spontaneous mutations as determined by similar experiments with unirradiated controls. It will be seen that the

curves for the two population types differ markedly in the first and second divisions, the number of mutant clones appearing in the first division being considerably smaller

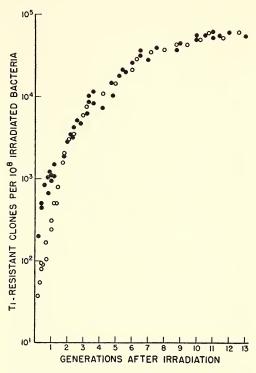


Fig. 1. Effect of nuclear multiplicity on delayed appearance of ultraviolet-induced T1-resistant mutants. Solid circles, uninucleate populations (24-hour cultures); open circles, multinucleate populations (late-lag-phase cultures). Ultraviolet dose, 800 ergs/mm². Survival, 10 per cent.

in the multinucleate than in the uninucleate population. By the end of the second generation, the number of mutant clones produced is the same for the two populations, and there is no further difference between them.

These results are in good agreement with expectation according to the hypothesis that the genetic determinants of phage resistance are located in the nuclear body. If phage resistance is a recessive character,

as seems likely on the basis of independent evidence, a multinucleate cell in which resistance to T1 had been induced in one of several nuclei would necessarily remain phenotypically sensitive at least until division and segregation could produce a clonal descendant containing only replicas of the mutated nucleus. For cells having four nuclei at the time of irradiation, two divisions would be required for complete segregation. Thus the observed difference between uninucleate and multinucleate populations is quite compatible with the assumption that the staining body designated as the nucleus is the carrier of the genetic determinants of phage resistance. Taken in conjunction with the evidence for the nuclear basis of lactose fermentation presented last year (Year Book No. 50), this constitutes additional support for the acceptance of the Robinow body as a true genetic nucleus in bacteria.

It should be pointed out, however, that these results might be explained on other grounds than those of nuclear segregation; and, although they are compatible with this hypothesis, they cannot be considered as proving it. The uninucleate and multinucleate populations differ in other respects than nuclear number, two of the major differences being cytoplasmic volume and length of lag phase before the first postirradiation division. Neither of these factors can be entirely ruled out as a possible source of the observed differences between the expression curves obtained for the two populations.

These results indicate also that segregation from heterocaryotic cells normally plays an insignificant role in the phenomenon of delayed appearance of induced mutants, since the majority of these are delayed far beyond the number of generations required for complete segregation.

Effects of Temperature on Spontaneous and Induced Mutation

Studies of the effects of temperature on spontaneous and induced mutation in *E. coli* were begun by Witkin and Fetherston. It is hoped that this work will yield information which will be useful not only in elucidating the nature of the delayed appearance of induced mutants but also in furthering our understanding of the mutation process in a broader sense.

Cultures of strain B/r were grown in synthetic medium with aeration for 24 hours, diluted in saline to a titer of 108 cells per milliliter, and irradiated with 800 ergs per square millimeter of ultraviolet light, with mechanical agitation during the exposure. Irradiation was carried out at room temperature. After irradiation, samples of the treated suspensions were plated on nutrient-agar plates and incubated at 37°, 25°, or 16° C. After various incubation times, sets of six plates were withdrawn, and were used to determine the total number of bacteria per plate and the number of T₁-resistant clones, by the washing and spraying technique described above in connection with the experiments on nuclear segregation. Expression curves, in which the yield of mutant clones was plotted against the number of postirradiation generations, were thus obtained for three different temperatures of incubation after irradiation with ultraviolet light. Control experiments using unirradiated cells were carried out in the same way, with incubations at the same three temperatures.

1. Effect of temperature on generation time. The rate of growth of untreated and irradiated bacteria on nutrient-agar plates was found to have a temperature coefficient of about 3. The generation time is 20 minutes at 37°, 1 hour at 25°, and 3 hours

and 20 minutes at 16°. The lag phase for untreated cells is 1 hour and 10 minutes at 37°, 3 hours and 30 minutes at 25°, and 10 hours and 40 minutes at 16°. For irradiated bacteria, the lag phases are 3 hours, 10 hours, and 30 hours at 37°, 25°, and 16°, respectively.

2. Effect of temperature on spontaneousmutation rate. Figure 2 shows the number

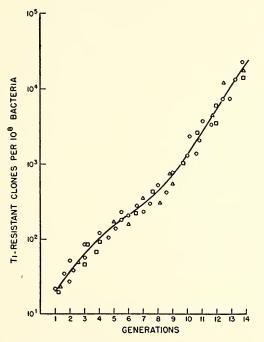


Fig. 2. Spontaneous mutations to Ti resistance. Circles, incubation at 37° C; squares, incubation at 25° C; triangles, incubation at 16° C.

of T1-resistant clones, per 10⁸ unirradiated cells plated, as a function of generations at 37°, 25°, and 16° C. The identity of the three curves shows that the process of spontaneous mutation to T1 resistance, expressed as the probability of mutation per bacterium per unit time, has the same response to temperature as does the generation time (Q_{10} ca. 3). The probability of mutation per bacterium per generation, which is the conventional way of express-

ing spontaneous-mutation rate in bacteria, thus remains quite constant over the temperature range investigated. The data for spontaneous mutations are plotted in this rather unorthodox fashion so as to show most directly the magnitude of the corrections used in the preparation of expression curves for induced mutation. The same data can be used to calculate mutation rate according to standard formulas.

- 3. Effect of temperature on induced mutation. The response to temperature of induced mutation to T1 resistance is best shown by expression curves in which the number of detectable mutant clones per 10⁸ irradiated cells is plotted against post-irradiation divisions at 37°, 25°, and 16°. These curves, corrected for spontaneous mutations, are shown in figure 3.
- a. Effect of temperature on the final yield of induced mutations: The curve for bacteria incubated at 37° levels off at about 56,000 mutant clones per 108 irradiated cells; at 16°, the end-point number is about 2800, or 5 per cent of the yield at 37°. Thus, a profound effect of temperature on the yield of induced mutations is indicated, but the data obtained at 25° show that this effect cannot be expressed as a simple temperature coefficient. Although the data for bacteria incubated at 25° are still rather few, there seems to be little, if any, reduction of the number of induced mutations at this temperature as compared with the yield at 37°. The temperature effect on the final yield of induced mutations is much greater between 25° and 16° than between 37° and 25°.
- b. Effect of temperature on the pattern of delayed appearance of induced mutants: Figure 4 shows percentages of the final yield of induced mutations as a function of postirradiation divisions, at 37° and at 16°. The identity of the two curves shows that,

despite the large difference in final yield of induced mutations at the two temperatures, the span of generations required for a given fraction of the total to manifest itself is not modified by temperature. This

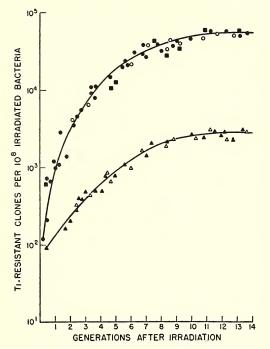


Fig. 3. Effect of temperature on ultravioletinduced T1-resistant mutants. Solid circles, incubation at 37° after irradiation; solid squares, incubation at 25° after irradiation; solid triangles, incubation at 16° after irradiation; open circles, first postirradiation generation at 37°, all others at 16°; open triangles, first postirradiation generation at 16°, all others at 37°. Ultraviolet dose, 800 ergs/mm². Survival, 10 per cent.

supports the conclusion of Demerec (this Year Book) that the pattern of delayed appearance is a characteristic and stable feature of a particular mutation.

c. The temperature-sensitive period: In the experiments described above, the irradiated cells were incubated at 37°, 25°, or 16° throughout the period of postirradiation growth. Another series of experiments was conducted in which the irra-

diated bacteria were incubated at 37° for one generation, after which the plates were transferred to 16° for the duration of the experiment. Reciprocal experiments were also made, in which the first division was at 16°, and the remaining divisions took place at 37°. The data of these experiments, also plotted in figure 3, show clearly

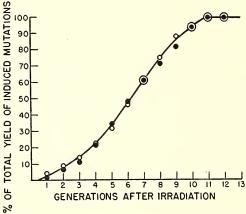


Fig. 4. Effect of temperature on pattern of delayed appearance of ultraviolet-induced Tiresistant mutants. Open circles, postirradiation growth at 37° C; solid circles, postirradiation growth at 16° C.

that the yield of induced mutations depends entirely on the temperature of incubation during the first postirradiation generation. If the first division proceeds at 16°, the final yield of mutations is 2800 per 10⁸ treated bacteria, whether the remaining divisions take place at 16° or at 37°; similarly, if the temperature during the first division is 37°, the end-point number is 56,000 whether the subsequent divisions are at 37° or at 16°. Although 10 to 12 generations are required to achieve complete manifestation of the end-point number, it is the temperature during the first division that determines the outcome.

In an attempt to define the temperaturesensitive period more precisely, irradiated cells were exposed to one temperature for

various fractions of the time required for the first generation, before the plates were transferred to the other temperature. Intermediate yields were obtained, the initial temperature exerting an effect proportional to the fraction of the first generation passed under its influence. At present, these results can be accounted for equally well by two hypotheses concerning the temperature-sensitive period: (1) that the critical period is confined to the actual division process, the intermediate yields being a result of mixtures of cells that have and have not completed the first division; or (2) that the temperature effect is exerted gradually throughout the entire lag phase and first division.

- d. Effect of temperature after the first postirradiation generation: It was pointed out above that if the irradiated cells are allowed to pass through the first division at a given temperature, the temperature during the subsequent generations has no effect on the course of the expression curve. Thus, after the first generation, the rate of appearance of mutant clones as a function of time has the same temperature coefficient as the processes of cell division and spontaneous mutation, about 3, so that the rate of appearance of mutant clones per generation is not affected by temperature.
- e. Dose-reduction equivalent of the temperature effect: The expression curve obtained at 16° after a dose of 800 ergs/mm² can be duplicated at 37° quite exactly when the dose is reduced to 100 ergs/mm².

Spontaneous-mutation rate, considered as the probability of mutation per unit time, was found in these studies to respond to temperature according to the Van't Hoff law with a Q_{10} of about 3. This agrees essentially with earlier work on spontaneous mutations in *Drosophila*. Another striking feature of these results, with regard to spontaneous mutations, is the

intimate relation revealed between mutation and cell division, strongly suggesting that the mutation process is associated with the mitotic cycle. These results are difficult to reconcile with those of Novick and Szilard, who, in studies using the chemostat, found that spontaneous mutations occurred at a constant rate per unit time, regardless of wide differences in generation time created by varying the availability of essential nutrilites.

The profound influence of postirradiation temperature on induced-mutation frequency may be taken as additional evidence that the mutagenic effect of ultraviolet radiation is indirect.

More than one concept of the induction and delayed appearance of mutations can be drawn from these experiments. Perhaps the best support is given to the hypothesis that ultraviolet light produces metastable genetic states, capable of stabilization at low temperature, and giving rise to the stable mutant form at a high rate during the divisions following treatment. Thus the delayed appearance of induced mutants would be a phenomenon akin to spontaneous mutation—a line of thought supported by the identity of the temperature responses of the two processes. Further temperature studies are planned to extend these investigations.

ZERO-POINT MUTANTS

Previous studies by Demerec and by Demerec and Latarjet (Year Book No. 45, 1945–1946, pp. 143–144) reported two classes of phage-resistant mutants induced by ultraviolet or X-ray treatment, namely, mutants that appeared among the treated bacteria before they had had a chance to divide (zero-point mutants), and those that continued to appear during several bacterial divisions after treatment (delayed mutants). Evidence accumulated in the

past two years, however, indicates that treated cells must divide before induced phage-resistant mutants can appear; in other words, that there are no zero-point mutants.

The first critical data on this point were obtained by Visconti during the winter of 1950–1951, when he studied the induction of phage T1–resistant mutants by ultraviolet radiation. Later his findings were substantiated by data obtained by Witkin in similar experiments with ultraviolet, by Demerec and Hanson in experiments using manganous chloride as the mutagen, and by Galinsky in studies of ultraviolet-induced resistance to phage T3.

It is now evident that the "zero-point" class of mutants does not exist, and that the earlier observations were not correct. It seems probable that in those experiments a fraction of the treated cells divided before the phage reached them and that thus mutants induced by the treatment had a chance to appear.

Tests of Chemicals for Mutagenicity

Among the several methods that we have developed for studying induction of mutations, the Sd-4 method is best adapted for determining whether an agent is mutagenic. This method was used successfully in our tests involving 52 chemicals (Year Book No. 49, 1949-1950, pp. 145-151). During the past year Demerec and Hanson have made such tests with eight compounds received from Professor A. Haddow, of the Chester Beatty Research Institute of the Royal Cancer Hospital, London, who uses them in studies of various problems of carcinogenesis. The object of our tests was to determine whether the Sd-4 method would be more efficient for determining mutagenicity than the Drosophila method now used by Haddow's group.

The following aromatic nitrogen mustard compounds were tested:

C.B.1044 (R44): NN-di-(2-chloroethyl)-p-toluidine

C.B.1045 (R45): NN-di-(2-chloroethyl)-p-anisidine C.B.1048 (R48): β-naphthyldi-(2-chloroethyl)-amine C.B.1181 (R181): 1:2:3:4-diepoxybutane C.B.1348 (R348): NN-di-(2-chloroethyl)-p-aminophenyl butyric acid C.B.2040 (GT40): 1:3-dimethanesulphonoxypropane

C.B.2040 (GT40): 1:3-dimethanesulphonoxypropane C.B.2041 (GT41): 1:4-dimethanesulphonoxybutane C.B.2058 (GT58): 1:4-dimethanesulphonoxybut-2-yne

The compound R181 is a liquid; all the others are solids, and only slightly soluble in water. Our procedure with the solid compounds was to shake an excess of the chemical in small bottles in distilled water for about an hour at room temperature, then decant the supernatant and use it in the experiments. Sd-4 bacteria, which had previously been washed in saline, were treated in various dilutions of the chemicals for 1 hour at 37° C and then plated on broth-agar medium containing no streptomycin. Since Sd-4 requires streptomycin for growth, the plated bacteria underwent only two or three divisions (residual growth) on the streptomycin-lacking medium. Any nondependent mutants, however, whether they had been present among the plated bacteria or originated during the period of residual growth, were able to form colonies. The mutagenic effectiveness of the treatment was determined by comparing the number of colonies obtained from treated bacteria with the number obtained in control platings.

The results of our experiments are summarized in table 2. R48 was the only compound that did not show mutagenicity. Since it is insoluble in water, however, it seems very likely that under our experimental conditions it was not able to penetrate the bacterial cell. The solubility of GT41 is very slight. Probably the reason more mutants resulted from the 1/16-dilution treatment than from any other dilution of this compound is that some par-

ticles were accidentally transferred with the supernatant when the dilutions were made and a disproportionate number of them were dissolved in the 1/16-dilution bottle. For all the other compounds, the data indicate that mutagenicity is correlated with concentration. These tests have shown that the Sd-4 method is effective in detecting mutagenicity in chemicals of this group when they are soluble in water. Since the method is very simple, and tests of a new chemical can be completed in about a week, it should be convenient for screening purposes.

TABLE 2

Results of mutagenicity tests using the SD-4 strain of Escherichia coli

(s%, per cent survivors; m, mutants per 108 survivors. Spontaneous mutation frequency is between 5 and 15 per 108.)

Concentration	R44		R45		R48		R181*		R348†		GT40‡		GT41		GT58	
	s%	m	s%	m	s%	m	s%	m	s%	m	s%	m	s%	m	s%	m
Saturated	55	122	24	71	44	18	0		clu	mped	0.22	2390	100	29	20	371
1/2	60	86	48	42	54	16	0		clu	mped	4.8	2624	94	22	28	293
1/4	67	45	37	35	55	13	3	522	3	2940	59	442	91	22	30	155
1/8	70	41	58	22	42	21	21	324	16	1167	53	254	98	9	36	103
1/16	66	41	45	18	52	18	42	178	30	429	54	145	100	125	43	50
1/32	66	28	51	16	37	32	42	101	39	519	52	73	82	13	46	47
1/64	90	21	56	12	40	13	55	52	44	247	50	53	95	13	43	32
1/128	82	17	48	15	51	20	50	39	45	131	60	34	96	6	59	23

* Liquid; first concentration is 1/100 dilution of the original.

† Dilutions are in steps of 1/5.

First concentration is 1/100 dilution of the original.

INITIAL STEPS IN THE REPRODUCTION OF BACTERIOPHAGE

A. D. HERSHEY AND MARTHA CHASE

The idea has developed during the past few years that intracellular viruses, bacterial and animal, differ in fundamental ways from the extracellular particles that have heretofore been subject to biochemical and biological study. This was most clearly shown by Doermann (Year Book No. 47, 1947-1948), who found that no infective virus particles could be recovered by bursting open bacteria during the first 10 minutes after infection with phage. Inasmuch as virus is undoubtedly multiplying in the cells at this time, the existence of a masked, vegetative structure is clearly implied. Information about this structure has come chiefly from genetic and biochemical study; and at present we are concentrating on the latter. Our results may be summarized as follows.

It is generally agreed that extracellular particles of phage T2 consist chiefly of protein and desoxypentose nucleic acid. We have used S35 and P32 as convenient labels for these two components. When radioactive phage is suspended in a three-molar solution of sodium chloride, and the suspension is diluted very rapidly with water, the phage particles are disrupted, as can be shown in several ways. First, as T. F. Anderson found, the infectivity of the particles is lost, and electron micrographs reveal empty tadpole-shaped ghosts. Secondly, as Herriott showed, the ghosts can be freed of phosphorus by treatment

with the enzyme desoxyribonuclease and centrifugal washing. The phosphorus-free particles are capable of attaching to phagesusceptible bacteria and lysing them. Finally, radiochemical analysis of osmotically shocked suspensions shows that nearly all the sulfur-containing protein of the phage can be adsorbed to bacteria, or precipitated with antiphage serum, leaving nearly all the phosphorus in solution. Analysis of these materials shows that phage particles consist of a protective membrane, chiefly protein, which is responsible for the specific attachment of virus to bacterium, and a nonantigenic core consisting chiefly or entirely of nucleic acid and a small amount of a free basic amino acid. The amino acid has not been identified, but is probably identical with the "fast arginine" found by Dent in extracts of various tissues.

The interaction of virus particles with phage-sensitive bacteria can also be studied by radiochemical and enzymatic methods. After phage attaches to heat-killed bacteria, the viral phosphorus can be digested out of the cells with desoxyribonuclease, leaving the viral sulfur behind. (Experiments of this type were first performed by A. F. Graham and collaborators.) Since whole phage is not affected by the enzyme, this result shows that the viral membrane is altered in some way by the attachment of phage to bacterial cells. A similar change can be demonstrated after the attachment of phage to live bacteria, but only if the infected cells are damaged in some way to permit the enzyme to get inside. This shows that the viral nucleic acid, at least, enters the cell on infection, and suggests the possibility that it is ejected from the viral membrane into the cell more or less as it is discharged into solution during osmotic shock.

The discharge of nucleic acid from the viral membrane following attachment to

bacteria was demonstrated in two ways. One consisted in allowing isotopically labeled phage to attach to broken fragments of bacterial cells, and then centrifuging the mixture to separate the cellular debris from soluble materials. It was found that nearly all the phage membranes (sulfur) sedimented with the debris, whereas about half the phage nucleic acid (phosphorus) remained in solution.

The second method of demonstrating the discharge of viral nucleic acid was suggested by the recent electron micrographs of T. F. Anderson, which show that the primary attachment of virus to bacterium takes place by means of the exceedingly slender tail of the virus particle. We found, using isotopically labeled phage, that this attachment could be broken by spinning a suspension of infected cells in a Waring Blendor. This treatment strips the viral membranes from the infected cells, and leaves the viral nucleic acid inside. What is most remarkable, the ability of the infected cells subsequently to yield viral progeny is not affected by stripping. The viral nucleic acid thus becomes the center of interest in the investigation of intracellular phage, though further work is required to give this statement chemical precision.

Another indication of the importance of nucleic acid in viral growth is the following. If bacteria are infected with isotopically labeled phage, and the viral progeny are subsequently isolated and analyzed, it is found that nearly half the nucleic acid phosphorus of the parental virus has been transferred to the progeny (Putnam and Kozloff). The sulfur-containing amino acids of the parental viral membranes, on the other hand, remain metabolically inert.

The initial steps in viral growth—adsorption of virus to bacteria and injection of viral nucleic acid into the cells—occur

independently of bacterial metabolism. The multiplication of virus, however, is tied to cell metabolism. As a first approximation, we may think of the initial phases of this multiplication in terms of the synthesis of the specific viral nucleic acid. Corollary ideas are that the viral nucleic acid functions as the sole agent of genetic continuity in the virus, and that the synthesis of viral protein (likewise a specific substance foreign to the uninfected cell) occurs de novo as a late step in viral growth. These ideas may prove correct, incorrect, or untestable, but they are likely to orient research on viral growth for some time to come.

A fortunate circumstance will make it possible to study the economy of nucleic acid synthesis in infected cells. We find

that bacterial nucleic acid contains one characteristic pyrimidine (cytosine) and viral nucleic acid another (unidentified) in addition to common constituents. The absence of cytosine from the nucleic acid of T2 had previously been reported by Marshak; and Cohen and Wyatt, independently of us, have also found the unidentified substance. We have developed a quantitative chromatographic method for analyzing mixtures of the two kinds of nucleic acid. Preliminary experiments show that the method can be applied to infected cells. It is to be hoped that this development may lead to a method for measuring the size of intracellular viral populations, and thus bring closer together genetical and biochemical questions about growth.

RECOMBINATION IN BACTERIOPHAGE

N. VISCONTI AND R. C. COON

Last year (Year Book No. 50, 1950–1951, pp. 194–195) we reported some genetic experiments with phage T2. These experiments have been extended, and a complete theory of genetic recombination in phage has been worked out, in collaboration with Professor M. Delbrück, of the California Institute of Technology. The publication presenting this theory is now in press.

Recombination has also been studied in relation to the growth cycle of phage in the bacterial cell. This part of the work was conducted in collaboration with Dr. C. Levinthal, of the University of Michigan, who was a guest at our laboratory for a period of one month. In the course of this work we discovered that infected bacteria are resistant to lysis from without.

Using acquired resistance to lysis from without as a technical device to superinfect bacteria with a very high multiplicity of phage particles, we were able to study in further detail the mutual exclusion between related phages already described by Dulbecco.

Apparent Negative Interference in Phage Crosses

Crosses between pairs of mutants of T2 reveal characteristics that simulate negative interference. Suppose that a and b are the recombination values between the first and second and between the second and third of three linked markers, and c is the recombination value between the first and the third. Then c=a+b-I2bc, where I is a coefficient of interference. In phage crosses, I is greater than 1. Another way of showing this apparent negative interference is the following. Suppose we measure the recombination value, a, between the first and second markers, and then repeat the measure, selecting the class of individuals that shows recombination between the second and third markers. In

a genetic cross, an increased recombination frequency resulting from this selection would be attributed to negative interference. An effect of this sort is characteristic of crosses with T2.

Since we already know that the mixed infection of a bacterium is not analogous to a genetic cross, negative interference should be considered in the light of a theory on the mechanism of recombination in phage. Measures were made using markers h, r2, and r7, belonging to the same linkage group, with equal and unequal multiplicity of the parental types. The data obtained agreed with the predictions of the theory of genetic recombination summarized later in this report.

GROWTH AND RECOMBINATION IN PHAGE

In phage crosses, the values for recombination between two different markers depend on the time of lysis. Doermann was the first to show that after premature lysis fewer recombinants are found than after normal lysis. On the other hand, Hershey noticed an increase in proportion of recombinants when lysis is delayed. In collaboration with Dr. Levinthal, we investigated this phenomenon.

In the first part of this study we were concerned with the general problem of lysis inhibition. Doermann had already established the fact that when infected bacteria readsorb more phage before lysis, lysis is delayed for some time. Instead of adding a second input of phage, we used the following technique. A broth culture of the sensitive bacteria was grown under aeration. When it reached a concentration of 2×107 bacteria per milliliter, the culture was centrifuged and resuspended in broth at a concentration of 1×108 per milliliter. Phage was added at a multiplicity of 10 and the culture incubated at 37° C under aeration. It was assumed that the first phage to be freed would reinfect the other bacteria, thus delaying their lysis. After different intervals of time the culture was titrated by dilution and plate counts. Two or three hours were required for complete clearing of the culture. Figure 5 shows the results of the titrations. Comparing this curve with the one-step growth curve obtained by diluting before lysis, we can only conclude that lysis started earlier and that final phage titer was approximately the same in the lysis-inhibited culture. Further experiments clarified this result, as follows:

- 1. Up to 60 minutes, if the lysis-inhibited culture is centrifuged at low speed, a visible pellet is obtained. On dilution and counting of the resuspended pellet and of the supernatant, it turns out that 90 per cent of the phage is to be found in the pellet. This shows that bacteria burst during dilution manipulations. Appreciable numbers of extracellular phage are not found until the sixtieth minute.
- 2. All the bacteria burst upon dilution. If the last dilution tube is kept at 37° C for 90 minutes, no increase in phage titer takes place.

These findings can be explained in the following way. From 15 minutes on, bacteria burst open when the suspension is diluted. Starting at about 20 minutes, bacteria begin to lyse in the culture, but the phage they yield is readsorbed and destroyed by the other bacteria. As time goes on, fewer bacteria remain to be lysed, but the total number of phage particles obtained is the same. This last fact must be interpreted as a coincidence resulting from the decrease in number of bacteria and the increase of phage yield per bacterium.

If, instead of infecting the bacteria at the beginning with one kind of phage, we use equal numbers of two mutant types, we obtain recombinants throughout the growth cycle of the phage in the bacterial cell. Parental types hr_{13} and $h^+r_{13}^+$ were used for this experiment. Two closely linked loci were chosen in order to avoid

THE MECHANISM OF GENETIC RECOMBINA-TION IN PHAGE

The general theory of recombination in phage is based on results obtained by

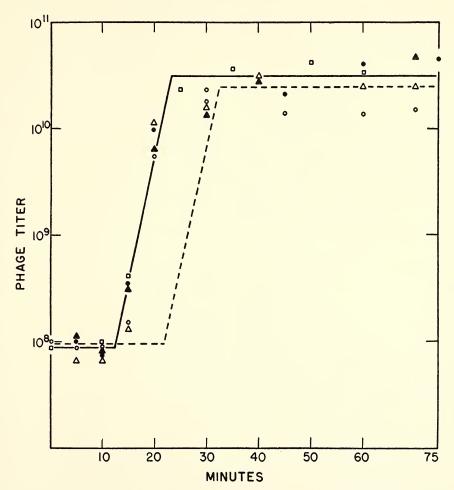


Fig. 5. Growth curve of a lysis-inhibited culture (solid line) and of a culture diluted before the end of the latent period (dashed line). Titers are expressed per milliliter of undiluted culture. Each of the symbols represents a separate experiment.

an approach to genetic equilibrium. The result of this experiment is shown in figure 6. The frequency of heterozygotes (2 per cent mottled plaques) remained constant throughout the experiment. The bearing of these results on the general theory of recombination is discussed in the next section.

Hershey and Rotman, and by Doermann, as well as on our results described above. The main assumptions of the theory are the following: (1) After infection, phage particles are transformed into noninfective particles (vegetative phage). (2) Vegetative phage multiplies up to the time of lysis. (3) Beginning at 10 minutes, vege-

tative phage is transformed into mature phage particles. (4) Mature phage particles within the bacterial cell do not revert to the vegetative condition, and do not mix genetically. (5) Mating is by pairs, and at random with respect to partner. The rate of mating, probably low at the similar to that responsible for Mendelian segregation in higher organisms.

RESISTANCE OF THE INFECTED BACTERIAL CELL TO LYSIS FROM WITHOUT

When a large number of phage particles is adsorbed to a bacterium, lysis is

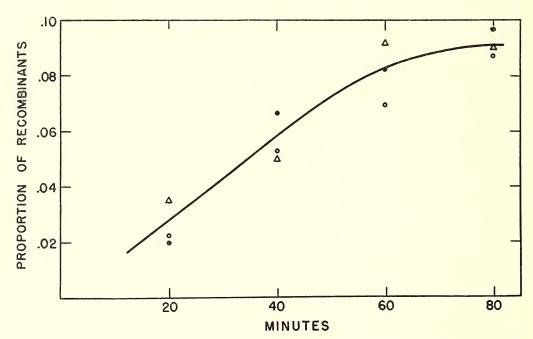


Fig. 6. Frequency of recombinants in the cross $hr_{13} \times h^+r_{13}^+$, plotted against time of dilution in a lysis-inhibited culture. Each of the symbols represents a separate experiment.

very beginning, becomes constant with time.

The genetics of phage thus becomes a problem of population genetics. The average number of matings per particle and the linkage values per mating must be calculated from the experimental data. For normal lysis at 22 minutes, the number of matings is around five. Mating goes on after this time if lysis is delayed. Linkage maps can be constructed that show recombination frequencies per mating. In these maps there is no evidence for interference. On the whole, it appears that the genetic mechanism operating in phages is

induced very rapidly. No new phage is liberated; on the contrary, the adsorbed phage is lost. Delbrück, in 1940, described this type of lysis as lysis from without (LFW), in contrast with lysis from within, which occurs, with liberation of phage, at the end of a characteristic latent period. In the same paper, Delbrück described some microscopic observations of LFW. Bacteria infected with a 200-fold excess of phage start swelling to an oval or spherical shape a few minutes after infection. The spherical structures persist for some time.

In some experiments done for other purposes, we came across the remarkable

fact that infected bacteria become resistant to LFW. This can be simply demonstrated by comparing a turbid growing culture infected with phage and a noninfected control. Five or six minutes after infection of one of the samples (with a multiplicity of between 5 and 10), we add concentrated phage (with a multiplicity of over 500) to both samples. Within a few minutes the control sample clears, while the turbidity of the superinfected sample remains unchanged.

Some quantitative methods have been worked out to measure acquired resistance to LFW. One method consists in infecting the bacteria with a mixture of r and r⁺ in such a way that every bacterium plated before burst will give a mottled plaque on the plate. At different intervals after the first infection, a second input of phage is added at a very high multiplicity. Five minutes later, antiserum is added to inactivate unadsorbed phage. After 4 additional minutes the culture is diluted and plated; only infected bacteria and free phage surviving the exposure to antiserum will give plaques. Free phage will give either r or r^+ plaques; infected bacteria will give mottled plaques. This method proved useful because it is difficult to get rid of all free phage, using antiserum, when the multiplicity is very high. By counting the mottled plaques, survival ratios of the bacteria can be calculated. If the interval between first and second infection is 1 minute, less than 1 per cent of the bacteria resist LFW. With an interval of 4 minutes, over 50 per cent of the bacteria survive.

A more accurate method is the following. A bacterial culture is infected in buffer with a multiplicity of o.i. After adsorption, free phage is eliminated by twice centrifuging and resuspending the bacteria in buffer. Then growth of the

phage is started by adding broth at 37° C. Samples of the culture are lysed at intervals by adding a large amount of ultraviolet-killed phage. After 5 minutes, during which LFW occurs, the culture is diluted and plated. Only surviving bacteria will give plaques. Survival curves can be determined very accurately by this method. Using a multiplicity of 1000 phage particles per bacterium to induce LFW, we obtained complete resistance 8 minutes after the first infection.

Acquired resistance to LFW is specific, in the sense that bacteria infected with T2, but not bacteria infected with T1, become resistant to LFW induced with phage T2.

Bacteria that are already infected adsorb phage at the normal rate. Resistance to LFW is not due to failure of adsorption.

MUTUAL EXCLUSION BETWEEN RELATED PHAGES

Dulbecco has shown that when bacteria infected with phage are superinfected with a related phage, the first phage excludes the second from the yield. Exclusion becomes more and more complete as the interval of time between the two infections increases. This phenomenon has been called mutual exclusion between related phages, to distinguish it from mutual exclusion between unrelated phages, which has to be explained on a different basis. In Dulbecco's experiments, the multiplicity was approximately 1, for both first and second infection. Bacteria were plated before burst, and, as r and r^+ strains were used for first and second infection, mixed bursts gave mottled plaques. Delaying the second infection greatly reduced the frequency of mottled plaques.

There are two possible explanations of this fact: (1) Some bacteria develop, later than others, an all-or-none exclusion effect. (2) As the interval of time between the two infections increases, there is a decrease in the probability that each adsorbed phage particle will grow. Only the second alternative leads to the prediction that the frequency of mottled plaques should increase with the multiplicity of the second infection.

Knowing that infected bacteria become resistant to LFW, we were able to increase greatly the multiplicity of the second infection. Our experimental results show very clearly that, with any interval of time between the two infections, the frequency

of mottled plaques is directly related to the multiplicity of the second infection. It was shown that the multiplicity of the first infection is also important in the exclusion phenomenon; increasing this multiplicity reduces the frequency of mixed bursts.

We can conclude that the infected cell undergoes some change which decreases the probability that subsequently adsorbed phage will take part in growth. This change is probably related to resistance to lysis from without, and to other changes described by other authors.

MUTABLE LOCI IN MAIZE

BARBARA McCLINTOCK

Study of the origin and nature of instability of genic action at a number of different known loci in maize chromosomes has been continued during the past year. At any one locus of known genic action, different types of instability expression can appear. A hypothesis has been developed to account for the origin and subsequent behavior of these various types of unstable condition. Each type is considered as reflecting the operation of a particular chromosomal system that controls the action of the genic components at the locus. These controlling systems appear to be composed of distinct chromosomal units; but, unlike the genes, these units may move from one location to another within the complement.

Two well defined classes of controlling system have been dealt with in our study. They have been termed single-unit systems and two-unit systems. In the single-unit systems, only one controlling unit is recognized. When such a unit moves to a new location in the chromosomal complement, it imposes its specific mode of control of action of the genic components at this location. The time and place of

genic action, as well as its type, will be an expression of the specificity of the particular controlling unit. Two-unit systems operate differently. When one of these units is inserted adjacent to a particular gene, it may immediately alter genic action, giving rise to a recognizable mutation. Changes in this unit may subsequently occur. These are reflected in altered expressions of the associated genic components. They occur only when the second unit of the two-unit system is also present in the nucleus. This second unit is independently located in the chromosomal complement. Instability of genic expression is a reflection, therefore, of the interaction of the two units. Changes in the gene-associated unit control the types of change in genic action, whereas the time and place of such changes are controlled by the second unit.

The main purpose of recent studies has been to examine the same system of instability expression at a number of different known loci, and to compare the action of various systems that may operate at the same locus.

The possibility of recognizing the opera-

tion of a particular system controlling instability of genic expression depends on its specificity. The Ac component of the twounit Ds-Ac controlling system, described in previous reports, is highly specific in its action. Its operation is readily detected. In plants having Ds and Ac, mutability under the control of Ac has appeared at several known loci. The first recognized case was designated c^{m-1} . The origin of c^{m-1} by transposition of Ds to the locus of C has been described in previous reports. Subsequently, the Ds-Ac system of mutational control appeared at the Bz and Wxloci. Furthermore, several independent inceptions of this controlling system have occurred at each of these three loci. Other systems of mutational control have arisen, however, at the same loci. Since instability of genic action is considered to be an expression of the operation of a particular controlling system, which has been incorporated at the locus of the gene concerned, and not an expression of changes in the gene itself, it is to be expected that any one system can operate at any locus of known genic action. This expectation could be tested; and the Ds-Ac two-unit controlling system was chosen for this purpose. Instability at a selected locus can arise whenever the Ds component is transposed to the locus. By technically simple methods, it should be possible to detect the presence of this system within a few cell generations after incorporation of Ds at the locus. Two loci considered particularly appropriate for the initial tests were selected: that of A_1 and that of A_2 . The results of these tests are summarized in the following section.

Origins of Instability at the A_1 and A_2 Loci

The genic components at the locus of A_1 in chromosome 3 and the locus of A_2

in chromosome 5 are associated with the development of anthocyanin pigment in the aleurone layer of the kernel and in the plant tissues. If both Ds and Ac are present in plants homozygous for these two dominant, stable factors, instability may arise at either locus if Ds is transposed to it. (By use of the term factor instead of gene, it is hoped that misconceptions regarding the nature of the change affecting the genic components at a locus may be avoided.) Transpositions of Ds usually occur late in the development of sporogenous tissues. Should one such event insert Ds at the locus of A_1 or A_2 in these plants, genic action at the affected locus might be altered. Such altered action could be detected in kernels on the ears of these plants if the progeny of the cell in which the transposition occurred produced a female gametophyte, and also if the sperm nucleus entering this female gametophyte carried the known recessive, a_1 or a_2 , whose action is unaffected by Ac. Color development in the aleurone layer of the kernel so produced might be modified. Variegation might be exhibited if Ac was also present in this kernel.

The plants used as the female parents, in crosses to test this theory, had Ds located in the long arm of chromosome 5, closely linked to the factor Pr. One Ac factor was present, and all plants were homozygous for the stable, dominant factors A_1 and A_2 . Pollen from plants homozygous for either a_1 or a_2 was placed on the silks of such plants. The resulting ears were examined for kernels showing variegation of aleurone color. Seventy-one ears were obtained from the first cross and 120 from the second. One kernel showing variegation for aleurone color was found on an ear when a_1 had been introduced by the male parent, and three were found on three different ears when a₂ had been introduced by the male parent. Plants were grown from all four kernels, and tests initiated to determine the nature of the instability expressed.

In the plant derived from the single variegated kernel appearing in the first cross, mutability was being expressed at the locus of A_1 . This new mutable condition, designated a_1^{m-4} , proved to be Accontrolled. It arose at the normal A_1 locus in the Ds-Ac-carrying female parent. This could be determined from linkage studies. The female parent had been homozygous for A_1 and Sh_2 , whereas the male parent had been homozygous for the recessive alleles. Sh_2 is very closely linked with A_1 . Close linkage of a_1^{m-4} and Sh_2 was evident in backcross tests. Three other independent inceptions of instability at this A_1 locus have been examined. Two of them, a_1^{m-1} and a_1^{m-2} , are not Ac-controlled; but the third, a_1^{m-3} , is Ac-controlled and arose in a plant having the very same constitution as that which produced a_1^{m-4} .

Plants were grown from the three variegated kernels derived from crosses in which the pollen parent had been homozygous for a_2 . In two of them, it could be shown that modifications had occurred at the locus of A_2 in the Ds-Ac-carrying parent. Both modifications resulted in instabilities of genic action at this locus, which were designated a_2^{m-3} and a_2^{m-4} . In the third plant, the chromosome 5 contributed by the female parent was not transmitted through either pollen or egg; therefore the nature of the alteration at A_2 could not be determined. Mutability at a_2^{m-4} is Ac-controlled. The nature of the controlling system associated with a_2^{m-3} has not yet been determined; it is not Accontrolled, however. Initial tests were complicated by the presence of a defect in the chromosome 5 carrying a_2^{m-3} , which prevented pollen transmission. The locus of the defect is closely linked with Pr. Transmission of a_2^{m-3} through the pollen was limited. It occurred only when the grain carried a chromosome 5 without the defect. Such a chromosome is obtained by crossing over between a_2^{m-3} and the locus of the defect.

In considering the origins of a_2^{m-3} and a_2^{m-4} , it should be recalled that Ds was present in the same chromosome that carried A_2 . In two of the three plants derived from the variegated kernels, the chromosome 5 with a modification at A_2 was also defective. It is known that events at Ds produce chromosomal alterations. Although some of them do not produce chromosomal aberrations that result in inviability, others do produce gross chromosomal defects, such as deficiency. It is not surprising, therefore, to find that some defect in chromosome 5, affecting viability, accompanied the inception of mutability in two of these three cases. This relationship affords additional, although indirect, evidence that altered genic expression originates through chromosome aberration.

Two earlier cases in our material of mutability produced by changes at the A_2 locus are designated a_2^{m-1} and a_2^{m-2} . Neither is Ac-controlled.

Instability of Sh_1 Action Induced by Ds

Two independent cases of insertion of Ds just to the left of the Sh locus in chromosome 9 have been studied. With regard to the hypothesis of origin of mutation through Ds events, these two cases have been exceptionally revealing. Early studies of Ds were focused on the phenomenon of its transposition from one location to another in the chromosome complement. For the sake of technical simplicity, cases were selected in which Ds had been transposed from one position in the short arm of chromosome 9 to another position within the same arm. Over twenty such cases

were examined in detail. In five of them, Ds had been inserted between I and Sh. These two factors lie close together in the chromosome, and approximately 4 per cent crossing over occurs between them. From linkage tests, it was determined that Ds had been inserted closer to I than to Sh in three of the five cases, and at approximately the same position in each. Crossing over between I and Ds was approximately one-fifth of that which occurs between I and Sh. In the other two cases, Ds was inserted just to the left of Sh. To determine crossover frequencies between Ds and Sh in these two cases, extensive tests were required. A number of kernels with exceptional phenotypes appeared in these tests. Studies were then initiated to determine the conditions associated with their appearance. They proved to be Dsinitiated mutations at the locus of Sh. A summary of the evidence may be given.

The initial tests were made in order to determine the crossover frequencies between I and Ds and between Ds and Sh. The tests were conducted as follows for both case I (Ds 4864A) and case 2 (Ds 5245): Plants having one or two Ac factors and carrying I, Ds, Sh, Bz, and Wxin one chromosome 9, and C, ds, sh, bz, and wx in the homologue, were crossed to plants homozygous for C, ds, sh, bz, and wx and having no Ac factor. From one such test with case 1, only a single kernel was found among a total of 4201 that could be considered to have a chromosome derived from crossing over between Ds and Sh. In phenotype, it was I sh bx wx; and Ds was present in the I-carrying chromosome. In 26 kernels, the phenotype was I sh Bz; and in 17 of them Ds was certainly present, located between I and Bz. (The presence of Ds may be detected only in those kernels that also have Ac.) In normal stocks, crossing over between Sh and Bz is approximately half that between I and Sh, or close to 2 per cent. If the kernels with the I sh Bz phenotype carried a chromosome derived from a double crossover in the two adjacent short segments, I to Sh and Sh to Bz, then kernels carrying the reciprocal crossover chromosome should also have been present. They should have had the C Sh bz phenotype. No kernels showing this phenotype were present. It would be even more difficult to explain in terms of crossing over the origin of the 17 kernels with Ds located between I and Bz. Double crossing over, involving the Ds-to-Sh and Sh-to-Bz segments, would have been required. This seems even less probable when it is recalled that only one kernel had a phenotype that could have appeared as a consequence of a single crossover between Ds and Sh. Obviously, the I sh Bz phenotype does not represent a product of crossing over. Another mechanism is responsible.

The results in similar tests of case 2 were much the same as those described for case 1. In one such test, 6683 kernels were obtained. Seven of them were I sh bz wx in phenotype, and Ds was present in the I chromosome. They could be interpreted as having originated through crossing over between Ds and Sh. Twentynine kernels were I sh Bz; and in 17 of them Ds was certainly present, located between I and Bz. No kernels having a C Sh bz phenotype appeared. The arguments given above for case 1, which excluded crossing over as a mechanism responsible for the appearance of sh in kernels having an I sh Bz phenotype, applied with equal force for case 2. On the other hand, this phenotype could be attributed to mutation at the locus of Sh. Since mutation to sh had not been observed to occur with such a high frequency in other genetic stocks of maize, or in those

cases in our material where Ds was inserted at other locations in the short arm of chromosome 9, it could be suspected that these mutations were produced by some event undergone by Ds. Tests were initiated to determine whether this was true.

Events at Ds will occur only if Ac is present in the nucleus. If the sh phenotype arises from a particular event at Ds, then no mutations to sh should occur if Ac is absent. Thus, had the experiment described above been conducted with plants having the same constitution with respect to markers in their chromosomes 9, but having no Ac factor, kernels with an I sh Bz phenotype would not have appeared on the ears, for no events at Ds would have occurred in any of the cells of these plants. Such a comparative test was conducted with case 2. All the plants in one culture had the same constitution as described above with respect to markers in chromosome 9, but differed with respect to Ac, which was present in some and absent from others. When those having Ac were crossed to plants homozygous for C, sh, bz, and wx, and carrying no Ac factor, some kernels with an I sh Bz phenotype appeared on the resulting ears, as in the experiment described earlier. When four of the sister plants that had no Ac were similarly crossed, no I sh Bz kernels appeared, among a total of 2553, on the resulting ears, and none of the kernels was variegated; for no Ac was present in any of them and therefore no breaks at Ds could occur. Pollen from these same four plants was placed on silks of plants homozygous for C, sh, bz, and wx and also carrying an Ac factor. Ds events could now occur, but only during the development of the kernels that had received Ds from the male parent and Ac from the female parent. Mutation to sh might originate in some of these kernels, which would

then show segments of the sh phenotype but would not be totally sh. Among 1677 kernels produced from this cross, none having I and Bz was totally sh in phenotype. In the reciprocal cross, 1213 kernels were obtained. In such a cross, the Bz phenotype can be detected only in those kernels that also have Ds and Ac. No I sh Bz phenotypes were found among them. It should be emphasized that in these reciprocal crosses no evidence of crossing over between Ds and Sh appeared; that is, in none of the kernels with an I sh bz wx phenotype was Ds detected. They apparently carried a chromosome derived from crossing over between I and Ds. In both case 1 and case 2, the frequency of crossing over between I and Ds seemed to be the same as that between I and Sh in other stocks. Similarly, no change in crossover frequency between Sh and Bz was noted in either case.

The comparative tests outlined above support the hypothesis that the *sh* phenotype in the *I sh Bz* kernels originates through events at *Ds*, when the latter is located just to the left of *Sh*; for this phenotype appears only when *Ac* is also present.

A second test of this hypothesis was conducted with both case I and case 2. In plants that are homozygous for I Ds Sh and carry an Ac factor, mutations to sh should occur in a few cells late in the development of the sporogenous tissue, or in cells of the gametophyte. Kernels that are I sh in phenotype should appear on the ears from reciprocal crosses with plants homozygous for C ds sh. The frequency of appearance of such kernels in these reciprocal crosses is recorded in table 3. Many of the sh kernels certainly carried Ds in the I chromosome. Its location between I and Bz could be affirmed in the majority of cases because of the pattern of variegation produced by breaks at Ds.

If the described mutation results from inhibition of Sh action by Ds itself, then reversion to Sh might result from a subsequent event at Ds that removed this inhibitory action. In order to obtain more evidence about the nature of the events that are responsible for mutations at this locus, plants were grown from some of the $I \ sh \ Bz$ kernels in which both Ds and Ac were present. Tests were conducted to determine the viability of the mutant sh when homozygous, the position of Ds, and whether or not reversion to Sh would occur.

TABLE 3 Frequency of mutation to $\it sh$ in reciprocal crosses

Cross	. I Ds	Sh/	C ds s	 h ♀
			by I D	
			I Ds S	
	by $C d$	s sh 3	1 Ac	♂
Kernel type	I Sh	I sh	I Sh	I sh
Case 1 (Ds 4864A)	644	7	6349	128
Case 2 (Ds 5245)	9082	18	5004	34

For case I, ten kernels were selected that had the I sh Bz phenotype and carried both Ds and Ac. Only six of them germinated. A mutant sh factor was present in four of the plants. It was not present in one plant, whose constitution was like that of the heterozygous parent: I Ds Sh Bz Wx/C ds sh bz wx; I Ac. The sixth plant was triploid, with the following chromosome-9 constitutions: I Ds Sh Bz Wx/I Ds sh Bz Wx/C ds sh bz wx. Two Ac factors were present. A mutant sh factor was present in one of these I-carrying chromosomes.

An ear derived by self-pollination was obtained from each of the four diploid plants having a mutant sh factor. The ratio of I to C kernels indicated no reduction in viability of the mutant when homozygous. In crosses of these four plants to those homozygous for C, ds, and bz, a

few kernels with an Sh phenotype appeared, but only on one of the test ears from crosses involving one of the four plants. It is impossible to be certain that they were not the result of contamination, but the associated phenotypes make it unlikely that they were. In three of the four plants, crossing over between I and Ds and Ds and Bz was the same as that which occurred in the heterozygous parent plants, indicating no decided shift in the position of Ds in the origin of the sh mutation. In the fourth plant, the frequency of crossing over between I and Ds was unchanged, but that between Ds and Bz appeared to be reduced.

In case 2, 21 kernels having an I sh Bz phenotype and also carrying Ds and Ac were selected for testing. Only 13 of them germinated. A mutant sh factor proved to be present in twelve plants. The constitution of the thirteenth plant was similar to that of the heterozygous parent: I Ds Sh Bz Wx/C ds sh bz wx: I Ac. The I-to-C ratio on ears derived from self-pollination of the twelve plants carrying a mutant sh factor indicated no reduction in viability of the kernels homozygous for this factor. Crossing over between I and Ds was unmodified in all twelve plants. That between Ds and Bz was unmodified in eight plants, but in four plants it appeared to be reduced. On the ears derived from crosses to plants homozygous for C, sh, bz, and wx, kernels having an I Sh Bz phenotype appeared in tests of three of the eight plants in which crossing over between Ds and Bz was unmodified. The rates of reversion were low in two plants, but markedly higher in the third plant (5933-1). This applied to rates of germinal mutation, that is, mutation occurring in the sporogenous or gametophytic cells. If mutation occurs during the development of the kernel, a sector with the Sh phenotype will be

formed. To be detected, such sectors must be large; in other words, the mutation must occur early in the development of the kernel. All late-occurring mutations will be overlooked. Some sectorial kernels developed on the ears derived from crosses of all three plants mentioned above, the frequency of such kernels being highest on the ears derived from crosses of plant 5933-1. It appeared that this plant carried a mutable sh locus; and tests conducted with the progeny of the plant during the winter of 1951-1952 fully confirmed its presence. Although Ac control of mutation is probable in this case, it cannot yet be demonstrated with certainty. Inability to detect all kernels showing mutation to Sh necessitates extended tests for this determination.

It is known from studies of Ds-initiated mutable loci involving factors associated with color development, which may be detected even in individual cells, that the frequency of reversion depends on the state of Ds. For some states, the rate is high; for others it is very low. Only three of the twenty examined Ds-initiated mutations to sh have provided certain evidence of subsequent reversion, although a fourth instance may possibly have occurred. The original I sh Bz kernels selected for testing did not show any detectable somatic reverson to Sh. They may represent, therefore, a selected class in which the state of Ds is one producing relatively few reversions. Tests of an unselected sample are now under way. They should give a better indication of the frequency of production of highly mutable sh loci through the medium of Ds events. Nevertheless, from the several tests outlined above, there appears to be little question that events at Ds, following its insertion just to the left of Sh, are capable of inducing mutation to sh. It is also certain that some of the mutants are capable of subsequent reversion to *Sh*. Since one of the selected *sh* mutants proved to be highly mutable, additional examples of high mutability may appear when appropriate methods for their selection are employed.

SUMMARY

Instability of genic action, under the control of the two-unit system of which Ac is one of the components, has been examined at six different loci. (See table 4.) Four of these six loci are associated with anthocyanin pigment formation (C, Bz, A_1 , and A_2), one with composition of the starch in pollen and endosperm (Wx), and the sixth with a morphological structure of the endosperm (Sh). It is clear that this twounit system may operate at loci concerned in quite different types of phenotypic expression. It cannot be stated that the system could operate at any known locus; but it should be capable of operating at many. Mutational behavior under the control of Ac always follows a distinct pattern, regardless of the locus involved. No mutations occur when Ac is absent. They appear only when Ac is present, and the time and place of their occurrence are an expression of the particular state and dose of Ac. At five of the above-mentioned loci (column 3 of table 4), other systems controlling genic expression are known to operate.

Some aspects of the behavior of a_1^{m-2} are unique in our studies. Two distinct classes of mutation occur. One class produces phenotypes indistinguishable from that given by the normal A_1 factor: deep pigmentation in the aleurone layer and intense pigmentation in all parts of the plant. The other class produces a complex series of changes affecting anthocyanin development in plant and aleurone. Mutations in the latter class form a graded series with respect to the intensity of pigmentation produced. In the plant, however, pigmen-

tation is always restricted to the root, stalk, sheath, auricle, and glume; none appears to be present in the leaf, except in the midrib and along the edge. The effects of a

TABLE 4

KNOWN LOCI (COLUMN I) AT WHICH MUTABILITY, UNDER THE CONTROL OF Ac, HAS ARISEN (COLUMN 2), AND MUTABILITY AT THE SAME LOCI CONTROLLED BY OTHER SYSTEMS (COLUMN 3)

(The figures following the symbols represent the sequence of appearance in the Cold Spring Harbor cultures.)

Symbol for normal, dominant factor at locus	Instability controlled by Ac	Instability controlled by system other than Ac
C	C^{m-1} C^{m-2} C^{m-4}	c^{m-3}
Sh_1	See text for cases	
Bz	$bz^{m-1} \\ bz^{m-2}$	bz^{m-3}
Wx	wx^{m-1} wx^{m-5} wx^{m-6}	wx^{m-2} wx^{m-3} wx^{m-4}
A_1	$a_1^{m-3} \\ a_1^{m-4}$	$a_1^{m-1} \\ a_1^{m-2}$
A 2	a_{2}^{m-4}	a_{2}^{m-1} a_{2}^{m-2} a_{2}^{m-3}

particular mutation of this class on intensity of pigmentation may not correspond in plant and aleurone. For example, a mutation producing very slight pigmentation in the aleurone may give very intense pigmentation in the affected plant tissues. Also, some of the mutations observed produced totally colorless kernels and intensely pigmented plants. Another distinctive feature of a_1^{m-2} is related to the effect

of crossing over in the immediate vicinity of this locus, that is, between a_1^{m-2} and Sh_2 . The majority of crossovers within this short segment result in mutation that appears to belong to the second class mentioned above. In contrast, tests of crossing over between Sh_2 and a_1^{m-1} , a_1^{m-3} , or a_1^{m-4} have given no such clear indication of mutation accompanying crossing over. Still other aspects of behavior peculiar to a_1^{m-2} have been observed, but description will be postponed until further evidence is available.

It is clear from our studies that different systems of control of genic expression can arise at any one locus in the chromosome complement, and that the same system may operate at different loci.

The organization of the chromosomes with respect to functional units, and the types of functional units that may be present, are not clearly understood. Our studies indicate that at least two classes of functional genetic unit are carried by the chromosomes: one of them potentially capable of determining a particular course of cellular reactions, the other associated with the control of this potential action. It has been possible to distinguish between these unit components at a locus and to describe, in terms of phenotypic expression, their modes of operation. Our studies also suggest that many mutations may be expressions of changes in controlling systems, the potential capacities of the gene units remaining unchanged. At present, there is no way to distinguish, on the basis of mutation alone, between alterations in potentialities of genic action and alterations in the controlling systems that leave the potential unchanged, except in those cases where the latter is clearly evident.

PATTERNS OF ORGANIZATION OF CELLULAR MATERIALS

B. P. Kaufmann, M. R. McDonald, H. Gay, E. C. Moore, G. Vanderbilt, and J. B. Larrabee

During the past year we have continued our cytochemical and biochemical analyses of patterns of organization of nuclear and cytoplasmic materials. Progress of these studies, in which cytologists and biochemists have worked in close collaboration, was again facilitated by a grant (RG-149) from the National Institutes of Health, U. S. Public Health Service. One project was carried out in co-operation with Dr. Kenneth Paigen, who was associated with the group as a Carnegie Institution Fellow. Other studies, reported below, were completed by Dr. Nebahat Yakar, who joined us in February 1951 as a Fellow of the Turkish Government and left in March 1952 to resume her teaching duties in the Department of Genetics and Pharmacobotany of Istanbul University.

Arrangements were made last fall with Drs. Thomas F. Anderson and Britton Chance, of the University of Pennsylvania, to use the electron microscope of the Johnson Foundation for Medical Physics in the analysis of salivary-gland chromosome structure. Techniques for spreading and photographing the chromosomes were developed by Miss Gay in collaboration with Dr. Anderson. Use of the electron microscope has greatly increased our opportunities for study of the finer structure of cell constituents.

Comparison of the Action of Trypsin and Pepsin

In efforts to establish the reliability of cytochemical methods employing enzymatic hydrolysis for identification and determination of patterns of association of nucleic acids and proteins, it was necessary to define precisely the course of events by

which each enzyme effects alteration of structure or modification of stainability of cellular materials. The need for such analysis in formulating an interpretation of the mode of action of an enzyme is illustrated by experiments in which salivary-gland chromosomes of Drosophila melanogaster were digested in pepsin or trypsin. Under the conditions commonly employed in cytochemical studies, solutions of trypsin effect dissolution of the chromosomes, whereas solutions of pepsin merely cause their shrinkage. On the basis of such observations it has generally been assumed that trypsin causes disintegration by digesting all classes of protein found in chromosomes, and that pepsin causes shrinkage by removal of a matrix protein, nonhistone in nature and unessential to the fundamental construction of the chromosome. The incorrectness of this interpretation of the degradational action of trypsin was revealed in experiments reported in Year Book No. 50 (1950-1951). They led to the conclusion that degradational changes effected by solutions of trypsin are due to swelling of partially degraded nucleoproteins in the presence of electrolytes as a consequence of the hydrolysis, but not the dissolution, of chromosomal proteins. Subsequent experiments have also shown that shrinkage in solutions of pepsin does not indicate selective action of this enzyme on one of two constituent classes of chromosomal protein.

It was found in these studies that either trypsin or pepsin could cause swelling or shrinking, depending on the method of treatment. For example, pepsin as ordinarily used, in 0.02 N hydrochloric acid at pH 1.4, causes shrinkage of salivarygland chromosomes, but if the shrunken

chromosomes are rinsed thoroughly in water and then immersed in 0.05 M potassium phosphate buffer and water, marked swelling occurs. Similarly, trypsin in buffer at pH 7.5 leads to swelling of the chromosomes, but if desoxyribonucleic acid (DNA) is first removed from the chromosomes, shrinkage occurs during subsequent treatment in buffered trypsin. Such experiments have shown that swelling and shrinking depend on the gel-like properties of the partially degraded nucleoproteins, and not on the selective action of either enzyme on any specific class of protein. Apparently trypsin, pepsin, and chymotrypsin (which we have also studied) attack peptide linkages in both histones and nonhistone proteins. In the course of cytochemical studies, in which treatments were given with two or more enzymes, it was observed that pepsin was often far more drastic than trypsin in its action on cellular substrates. Such results might have been anticipated on the basis of chemical studies reported by other workers, in which pepsin appeared to have a considerably wider range of specificity than trypsin.

CHROMONEMATA OF SALIVARY-GLAND CHROMOSOMES

The appearance of bands along the salivary-gland chromosomes depends, according to the most widely accepted interpretation, on the propinquity or confluence of homologous chromomeres of parallel-lying chromonemata. The longitudinally disposed chromonemata are not clearly defined in the chromosomes of *D. melanogaster* with customary methods of fixation in acetic acid and staining in aceto-orcein; and some workers have contended that the microscopically visible structural pattern, which suggests the presence of chromonemata, is a fixation artifact, resulting

from the distribution of chromatic material over the surfaces of achromatic droplets. According to this interpretation the true chromonemata are numerous and submicroscopic. In an effort to shed new light on this problem, salivary-gland chromosomes of *D. melanogaster* were treated with enzymes. In trypsin-hydrolyzed preparations chromosomes were observed that afforded striking evidence of the existence of chromonematic fibers connecting the chromomeres of adjacent bands.

Photographs of such treated chromosomes are shown in plate 1. These chromosomes were fixed and squashed in 45 per cent acetic acid, rinsed thoroughly in water, and then hydrolyzed for 8 minutes in trypsin in phosphate buffer at pH 5.5. The larger figure (pl. 1A) illustrates the over-all fibrous appearance of these partially swollen chromosomes; the two insets demonstrate the continuity of the chromonemata. Inspection of these photographs and the preparations from which they were made suggests that the chromonemata are peripherally disposed, and that the densely staining chromomeres constitute hoops or bands rather than disks. It is also apparent that the number of visible chromonemata or chromonematic aggregates is small (approximately 16). Although there seems to be a surface pattern of chromomeres and chromonemata, it cannot readily be explained on the basis of fixation artifact. The chromonemata are clearly defined and regularly disposed, and can be followed from chromomere to chromomere of adjacent bands for long distances along the chromosomes. In some chromosomes the chromonemata are associated regularly in pairs (lower inset); in others they appear to be single (upper inset). A possible explanation of the apparent surface pattern is that each of the homologues comprising the giant salivarygland chromosome is composed of a relatively small number of subsidiary units, which swell and become confluent in the course of development so that the orceinstainable material becomes peripherally disposed. Technical procedures, such as treatment with enzymes, that would alter the state of dispersion or aggregation of materials within the swollen cylinders might be expected to disclose such patterns as are seen in plate 1. In an effort to shed further light on this problem of submicroscopic organization, we have turned to electron microscopy.

A valid basis for critical comparison of electron micrographs with images obtained with the light microscope was ensured by using smears of whole chromosomes of D. melanogaster. The relatively large diameters of these chromosomes (3-5 µ), and their opacity to the electron beam under normal conditions, required the development of special techniques for their study. These techniques involved, first, adaptation of the critical-point method of T. F. Anderson, in whose laboratory the work was done, so that dehydration could be accomplished without surface-tension distortions; and, second, utilization of enzymes to improve the degree of contrast between various parts of the chromosomes.

This preliminary report of work done during the past eight months is confined to observations made on undigested chromosomes. A technique for smearing salivary glands of *Drosophila* larvae directly

on a Formvar membrane was devised. The glands were dissected in Ringer's solution, fixed in 45 per cent acetic acid, and squashed in 10 per cent lactic acid between a cover slip and a Formvar-coated slide. The cover was removed and the Formvar film floated off the slide onto water. A thin copper screen was then placed over the smear. To prevent drying in air, both film and screen were immediately picked up on a metal holder and passed through a graded series of alcohols. The tissue was desiccated by Anderson's critical-point method, which involves passage through amyl acetate and liquid carbon dioxide and subsequent vaporization of the carbon dioxide.

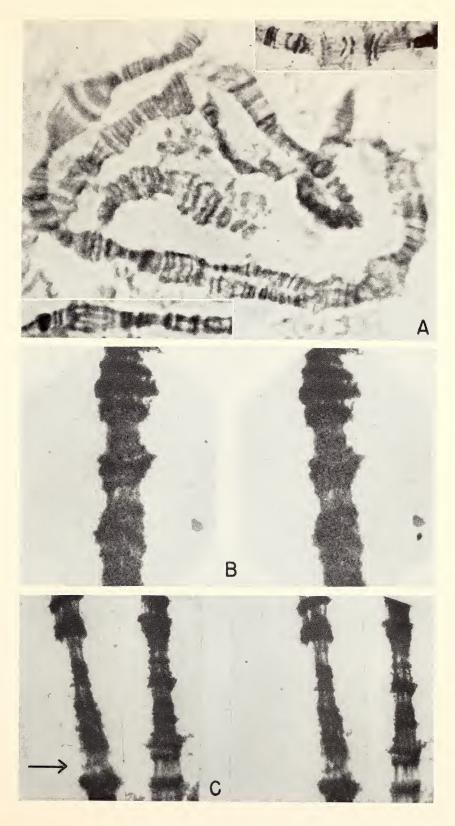
Stereo-pair electron micrographs of smeared chromosomes prepared in this manner indicate that the critical-point method preserves the chromosomes without flattening or distortion, and thereby provides good resolution. Several preparations of this type showed clear longitudinal fibers, especially in the interband regions. In several instances, the strands could be traced between the chromomeres of two adjacent bands (pl. 1B). The stereo views of the chromosomes suggest that in many bands chromomeres are located peripherally (pl. 1C). Unfortunately, there was considerable variation in the quality of the smear preparations, some being too dense to show any structure in the chromosomes. A further study of the technique to determine the reasons for this variability is

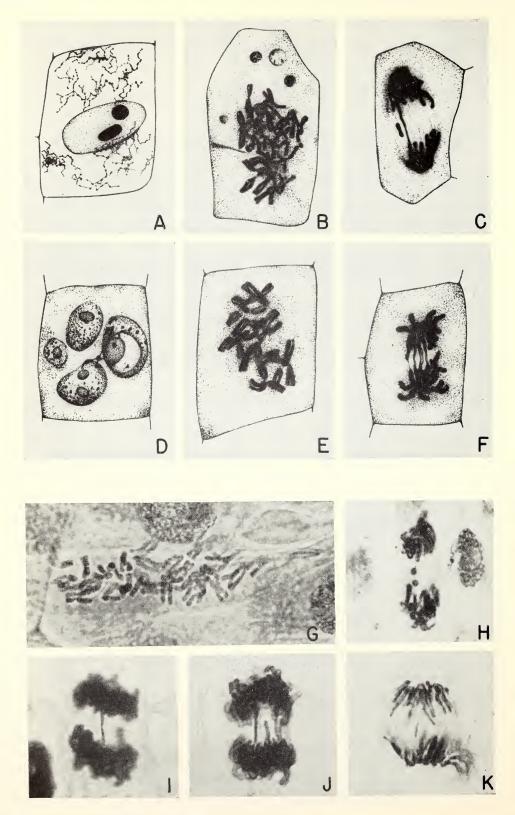
PLATE I

Chromonemata of salivary-gland chromosomes of *Drosophila melanogaster*

A. Phase-contrast photographs of smears hydrolyzed in trypsin and stained in aceto-orcein. The larger figure shows the over-all fibrous appearance of the chromosomes; the insets at upper right and lower left demonstrate the continuity of the constituent chromonemata.

B, C. Electron micrographs of smears prepared by the critical-point method. Stereo pairs (to be viewed with a stereoscope). B, longitudinal strands; × ca.5000. C, longitudinal fibers connecting chromomeres (see arrow); × ca.2000.





necessary. For comparative purposes, it will also be desirable to identify specific chromosomes, so that regions well known from the light-microscope studies can be analyzed. These aims will be pursued further with the smear techniques, using chromosomes isolated by micro dissection, or sectioned material if necessary.

Cytochemical Studies of Chemically Treated Cells

Dislocation of materials in pycnotic cells. The discovery about fifteen years ago, by A. F. Blakeslee and his colleagues in this Department, of the polyploidizing action of colchicine stimulated extensive studies of the effect of various chemical substances-including alkaloids, phenols, and heterocyclic compounds—on meristematic cells of plants. From these studies extensive data have been accumulated with respect to the structural changes induced. Relatively little has been discovered, however, about the chemical changes occurring in the cellular proteins and nucleic acids. In an effort to increase knowledge along these lines, cytochemical studies were undertaken this year to determine the nature of the chemical changes induced in root-tip cells of onion, lily, and broad bean by such chemicals as colchicine, caffeine, hydroquinone, pyrogallol, pyrocatechol, p-aminodimethylaniline monohydrochloride, 8-hydroxyquinoline, and tetrachloro-p-benzo-quinone (chloranil). The experiments were carried out by Dr. Nebahat Yakar during her stay at this Department as a Fellow of the Turkish Government.

Various types of aberration were produced by the chemicals tested. When used for short periods of time in low concentrations they produced chromosomal aberrations and mitotic disturbances. When used in higher concentrations or for longer periods of time they caused pycnosis, with characteristic agglutination of chromatin. The cytochemical studies reported here were concerned primarily with such pycnotic cells, since preliminary analysis had shown that there is no essential difference between cells of chemically treated and untreated roots, with respect to stainability of nucleic acids and proteins, unless the chemical has caused agglutination of chromosomal materials. In these studies, chemically treated roots and untreated controls were fixed, embedded in paraffin, and sectioned. Some of the sections were digested, before staining, in solutions of crystalline enzymes such as ribonuclease, desoxyribonuclease, and pepsin. These digested tissues, and those of the undigested controls, were stained in methyl green,

PLATE 2

Mitotic disturbances produced by chemical treatment of root tips: A–D, in broad bean; E–K, in onion. A–F produced by chloranil; G–K by ribonuclease. A–F, drawings by Nebahat Yakar; G–K, photographs.

A. Strands of methyl green-stainable desoxy-ribonucleic acid in cytosome.

B. Aneuploid giant cell with more than the diploid number of chromosomes and with 4 micronuclei.

C. Chromosome bridge and lagging chromosome (probably a fragment).

D. Trinucleate giant cell.

E. Scattered c-pairs resulting from failure of spindle activity.

F. Chromosome bridges joining the two anaphase groups.

G. Polyploid cell with ca. 32 c-pairs of chromosomes.

H. Chromosome fragments between separating anaphase groups.

I-K. Chromosome bridges and lagging chromosomes.

pyronin, methyl green-pyronin, fast green, or the Feulgen reagent.

In some of the pycnotic cells the toxic agent effected an over-all reduction of basophilia of both cytoplasm and nucleus, and filaments containing desoxyribonucleic acid (DNA) were observed in the cytoplasm (pl. 2A). A comparison of staining reactions with methyl green, methyl greenpyronin, and the Feulgen reagent in desoxyribonuclease-hydrolyzed and unhydrolyzed sections indicated that the cytoplasmic DNA was polymerized. These findings suggest that DNA escapes from the nuclei in the course of pycnosis; but whether it is removed as a high polymer, or as a low polymer which is then repolymerized in the cytoplasm, is not known.

In general, however, pycnotic nuclei were characterized by shrinkage and by intense, uniform staining, with loss of individuality of chromosomes. When stained with methyl green-pyronin, the chromatin of these cells appeared violet, and there were pools of deep-red-staining material in the nucleoli, around the nuclei, and around the condensed chromosomes. Experiments in which the nucleases and pepsin were employed indicated that the redstaining materials consisted of both ribonucleic acid and protein. The analysis also indicated that the violet color of the nuclei was probably due to the presence in agglutinated chromatin of both polymerized and depolymerized DNA.

Mitotic disturbances caused by chloranil. In the course of the studies outlined in the preceding paragraphs, marked mitotic disturbances were observed in sections of the broad-bean root tips being used as controls (pl. 2B, C, D). The root tips had germinated from seeds that had been coated before purchase with a fine, yellowish powder, which was found on inquiry to contain chloranil. Since this powder is widely

used as a fungicide, it seemed desirable to determine the effect of filtrates and suspensions of chloranil on root-tip mitoses. The filtrates of chloranil did not produce any abnormalities in treated onion roots; but suspensions of the chemical, and suspensions or filtrates of the chloranil-containing commercial preparation, induced various types of disturbance affecting chromosome form, number, and distribution (pl. 2E, F).

It has generally been assumed that commercial fungicides containing chloranil have no deleterious effect on treated seeds. The present findings indicate, however, that this chemical may act as a polyploidizing agent, or may induce cellular destruction by fragmenting or agglutinating the chromosomes.

Ribonuclease-induced chromosomal aberrations. The discovery reported above, that various chemicals which cause mitotic disturbances also effect dislocation of chromosomal nucleic acids and proteins, raised the question whether hydrolysis of these constituent materials by nucleases or proteases would produce chromosomal abnormalities. It seemed desirable to determine in the first place the action of ribonuclease, which has a molecular weight of about 15,000 and which might be expected to penetrate the living cell more freely than the larger molecules of other available nucleases and proteases. Results of the first preliminary experiments have just been obtained at the time of writing this report. Smears of some onion root tips that have been immersed in solutions of ribonuclease show chromosomal aberrations and mitotic disturbances. Some of the cells are polyploid, with scattered pairs of chromosomes that presumably have been produced as a consequence of failure of spindle formation (pl. 2G). Other cells contain chromosome fragments (pl. 2H, K); and in others

the separating ends of anaphase and telophase chromosomes have adhered to form chromosomal bridges (pl. 2I, J). No comparable mitotic disturbances have been observed in any of the control preparations. These experiments must be repeated with more extensive controls before the direct action of ribonuclease can be established with certainty; but if treatments with crystalline enzymes prove a reliable method for the production of chromosomal aberrations comparable to those induced by X-rays and various chemicals, we shall have a more critical basis than heretofore for appraisal of the processes involved.

Changes in quantity of nucleic acids in irradiated cells. A discussion was presented in Year Book No. 50 of the nature of the changes induced in cytoplasmic and chromosomal nucleic acids by ionizing radiations. After X-ray treatment of onion root tips, grasshopper embryos, and salamander larvae, increased concentrations of ribose nucleic acid (RNA) were detected in both chromosomes and cytoplasm of cells of the irradiated tissue, as compared with those of the nonirradiated controls. Such increases had been detected in the cytoplasm by other workers and attributed in some cases to inhibition of DNA synthesis and a concomitant accumulation of ribose nucleotides in the cell. It seemed essential to appraise this interpretation by determining whether irradiation results in compensatory adjustments in the levels of the two nucleic acids. For this purpose Paigen and B. N. Kaufmann carried out a study of the effect of wholebody X-ray irradiation on the nucleic acids of mouse liver. The results will only be outlined here, as a more complete report is presented by Paigen elsewhere in this volume.

It was found that after irradiation there is an initial drop in the levels of both RNA

and DNA, followed by a recovery. A second drop occurs about 18 hours after irradiation, and this is followed by a slow recovery to levels well above the control values. Similar changes were detected in the acid-soluble components of the tissue extracts obtained from mice killed 18 and 48 hours after irradiation. Cytochemical analysis of tissue sections by spectrophotometric methods confirmed the chemical findings concerning the drop in the level of DNA 18 hours after irradiation. These findings therefore afford evidence that the reactions affected by whole-body X-ray treatment do not involve the interconversion of nucleic acids and free nucleotides.

Effects of X-Radiation on Dilute Solutions of Crystalline Chymotrypsin

Results of studies on the inactivation of dilute solutions of crystalline trypsin by X-radiation were reported in Year Books Nos. 48, 49, and 50. It was noted that the number of micromoles of trypsin inactivated per liter per 1000 r (reaction yield) increased markedly as the concentration of enzyme was increased from 1×10^{-7} to 2×10^{-4} M. These results differed from those obtained with carboxypeptidase (the only other enzyme that had been studied extensively over a wide range of concentrations) by Dale and his co-workers, who reported constant reaction vields with concentrations between 6×10^{-6} and 6×10^{-3} M, although smaller reaction yields were noted at lower concentrations. During the past year, therefore, a brief study was made of the effect of X-radiation on dilute solutions of crystalline chymotrypsin (a third proteolytic enzyme prepared from beef pancreas), to determine whether its inactivation followed the concentration-reaction yield pattern characteristic of trypsin or that of

carboxypeptidase. The results of this investigation are summarized in table 5. It is evident that the reaction yield increased

TABLE 5

Inactivation of chymotrypsin by X-radiation: EFFECT OF ENZYME CONCENTRATION ON REACTION YIELD

(Solvent, 0.0015 N sulfuric acid; pH of solutions, 3.1; temperature, 3°-5° C)

Initial molar concentration of	Micromoles chymotrypsin destroyed per liter per 1000 r			
chymotrypsin	Mean	S. E.		
2.82×10 ⁻⁶	0.222	0.006		
2.94×10^{-6}	0.213	0.005		
4.52×10 ⁻⁶	0.247	0.006		
1.16×10^{-5}	0.253	0.006		
2.50×10^{-5}	0.255	0.008		
4.09×10 ⁻⁵	0.293	0.014		
1.22×10 ⁻⁴	0.385	0.061		
1.23×10 ⁻⁴	0.365	0.040		

as the initial concentration of chymotrypsin was increased. The increments were less marked, however, than in the trypsin experiments.

It was noted in Year Books Nos. 49 and 50 that the reaction yields for the inactivation of very dilute solutions of crystalline trypsin were lower when hydrochloric acid was used as the solvent than when sulfuric acid was used, owing probably to the affinity of the chloride ion for the component in X-rayed water responsible for the inactivation. Similar results have been obtained with chymotrypsin. For example, when the initial concentration of chymotrypsin was 2.9×10^{-6} M, the reaction yield with 0.0015 N sulfuric acid as the solvent was 0.213 \pm 0.005, whereas with 0.0015 N hydrochloric acid it was 0.149 \pm 0.004.

As with trypsin, the reaction yield for the inactivation of chymotrypsin by Xradiation was found to be a function of pH. The data are summarized in table 6. The results are similar to those obtained in other laboratories for the inactivation of chymotrypsin by ultraviolet radiation.

TABLE 6

Effect of pH on reaction yields for the inactivation of chymotrypsin by X-radiation

(Initial concentration of chymotrypsin, 2.7×10⁻⁶ M)

pH*	Micromoles chymotrypsin destroyed per liter per 1000 r			
	Mean	S. E.		
1.9	0.243	0.006		
2.8	0.206	0.005		
3.5	0.161	0.005		
4.9	0.138	0.005		
6.4	0.113	0.006		
9.3	0.144	0.013		

^{*} pH adjusted with sulfuric acid or sodium hydroxide.

EFFECT OF HYDROXYL RADICALS AND HYDROGEN PEROXIDE ON DILUTE SOLUTIONS
OF CRYSTALLINE TRYPSIN

The mechanism of the inactivation of dilute solutions of crystalline trypsin by X-radiation is essentially indirect, inactivation being brought about by an interaction between the trypsin molecules and the active entities produced by ionization of water. The nature of the active ingredient is unknown. Among the primary radicals and secondary products resulting from the ionization of water that might be involved are hydroxyl radicals and hydrogen peroxide. Experiments were made during the past year to test the effect of these chemicals on trypsin. Concentrations of hydrogen peroxide greater than o.oor M were found to interfere markedly with the method of assay used. Hence it was difficult to draw definite conclusions concerning the effect of hydrogen

peroxide on trypsin solutions. It was definitely established, however, that if hydrogen peroxide does inactivate trypsin, the inactivation is reversible on dilution. Since solutions of trypsin inactivated by X-radiation do not recover their activity on dilution, it is improbable that hydrogen peroxide is the component of "activated" water that is responsible for the inactivation of the enzyme. This conclusion was confirmed in experiments in which trypsin solutions were exposed to X-rays in the absence of atmospheric oxygen. Under these conditions the amount of hydrogen peroxide produced by the action of X-rays on pure water is practically negligible; yet the results did not differ significantly from those obtained in the presence of air. Solutions of trypsin exposed to hydroxyl radicals, generated either by the interaction of hydrogen peroxide and ferrous sulfate or by the action of ultraviolet light on hydrogen peroxide, were found to be inactivated. This inactivation, like that produced by X-rays, was not reversible on dilution. Furthermore, the rate of inactivation was an exponential function of the time of exposure. Hydroxyl radicals may, therefore, be among the substances responsible for the inactivation of solutions of crystalline trypsin by X-radiation.

DESOXYRIBONUCLEASE IN ONION ROOT TIPS

It was reported in Year Book No. 50 that when sections of onion root tips were

treated with certain samples of ribonuclease for long periods of time at 60° C, their stainability by the Feulgen procedure was markedly reduced. On the other hand, these samples of ribonuclease had no effect on isolated DNA, for which the Feulgen test is regarded as specific. Experiments conducted during the past year to determine the reasons for this discrepancy have established the following facts. Onion root tips contain an enzyme (desoxyribonuclease) capable of rapidly depolymerizing isolated DNA without the formation of inorganic phosphorus. This enzyme does not depolymerize intracellular DNA, however, unless the cells have been treated previously with ribonuclease. The rate of depolymerization is greatest at 60°-70° C, and at pH 5.5 or 6.0, depending on the solvent in which the reaction is carried out. Measurable amounts of this desoxyribonuclease still persist in root tips that have been fixed in Carnoy's fluid, stored in 70 per cent alcohol (for periods up to 3 years), embedded, and sectioned. The loss in Feulgen stainability of onion root tips after treatment with certain samples of ribonuclease is therefore probably due to the secondary action of residual root-tip desoxyribonuclease. Why treatment of onion root tips with other samples of ribonuclease does not reduce the intensity of the Feulgen reaction has not yet been established. Preliminary experiments suggest that they contain an inhibitor of the roottip desoxyribonuclease.

CYTOPLASMIC ORGANIZATION AT THE BIOCHEMICAL LEVEL

KENNETH PAIGEN, BERWIND N. KAUFMANN, AND REBECCA WOOD

X-RAYS AND NUCLEIC ACIDS

One of the most striking of the biological effects of ionizing radiations is that produced on the hereditary materials of the cell. In view of present concepts regard-

ing the role of nucleic acids in genetic mechanisms, it is important to understand the effects of ionizing radiations on nucleic acids.

For these reasons a study has been made,

using chemical methods, of the effects of whole-body X-ray irradiation on the nucleic acids of mouse liver. Observations were made on the nucleic acids at various times after irradiation. Two lines of experimentation were followed. In the first, the levels of nucleic acids of both types present after irradiation were studied quantitatively, and in the second the nucleic acids present at selected times after irradiation were isolated and characterized by quantitative determinations of the purine and pyrimidine bases present. This study was aided by a research grant (RG-149) from the National Institutes of Health, U. S. Public Health Service, in support of research by B. P. Kaufmann and McDonald.

In the first type of experiment, young adult male mice from a highly inbred line of the C58 strain were used. These mice were generously supplied by Dr. Mac-Dowell. Twelve were saved as controls, and sixty irradiated with 600 r of hard X-rays after a 24-hour starvation period. They were killed in groups of twelve at 6, 12, 18, 24, and 48 hours after irradiation. The nucleic acids were extracted from the excised livers essentially according to the method of Schneider. In addition to the nucleic acid extract, an "acid-soluble" fraction was obtained which contained any nucleic acid components of low molecular weight. The quantities of pentose nucleic acid (PNA) and desoxypentose nucleic acid (DNA) present in the nucleic acid extract were determined by colorimetric reactions for their respective sugars; and measurements of the total ultravioletabsorbing material present, in both the nucleic acid and acid-soluble extracts, were made with a Beckman spectrophotometer.

A summary of the results is shown in figure 7, in which the various quantities (expressed as percentages of the controls)

are plotted as a function of time after irradiation. The fact that the ultraviolet absorption of the nucleic acid extract (at $264 \text{ m}\mu$), which depends upon changes in the quantities of the nitrogenous bases, was in agreement with the colorimetric determinations of DNA and PNA, based on the sugar components, provides an independent check of the validity of the observations.

The most striking thing about these results is the parallel behavior of DNA and PNA, which is strongly suggestive of common or similar steps in the metabolism of the two nucleic acids. Such parallel behavior is strong evidence against the hypothesis advanced by some workers that irradiation results in a transformation of nucleic acid from one type to another.

It is to be expected that the exact time intervals at which the increases or decreases shown in figure 7 occur will depend both on the particular tissue studied and on the dosage level employed. The finding that whether the nucleic acid level observed in an irradiated animal is higher or lower than that of the control depends on the time after irradiation at which the measurements are made probably explains many of the conflicting reports in the literature. Previous workers, making measurements at a single time after irradiation, have arrived at different conclusions about the basic question whether irradiation results in an increase or decrease in the nucleic acids of a tissue.

Examination of the absorption spectra of both the nucleic acid and the acid-soluble extracts led to the conclusion that the only ultraviolet-absorbing materials that contributed significantly to the light absorption of these extracts were nucleic acids and their components. In view of this, it is very interesting that the fluctuations of the acid-soluble curve in figure 7 follow those of the nucleic acid curves. If

radiation-sensitive reactions were involved in the interconversion of nucleic acids and nucleotides, it would be expected that the levels of nucleic acids and nucleotides would vary reciprocally, since substrates of constituents by other tissues of the body. In the latter case the liver would be functioning as a nucleic acid "reservoir."

In the second type of experiment in this study, attention was focused on the ques-

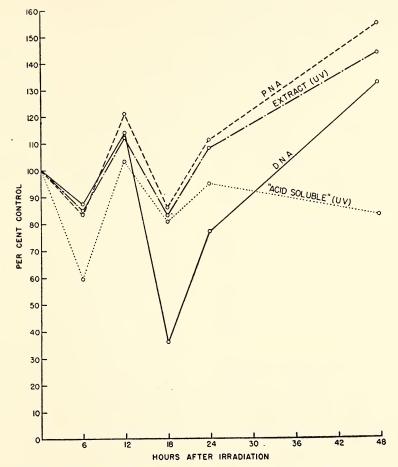


Fig. 7. Changes in the relative amounts of nucleic acid components of mouse liver during the postirradiation period. (X-ray irradiation.)

a reaction tend to accumulate as their conversion to a product is inhibited. The fact that the ultraviolet absorptions of the two extracts vary similarly may be interpreted in two ways. The radiation-sensitive reaction may precede the formation of nucleotides or other ultraviolet-absorbing materials; or the changes in liver may reflect the release or modification of nucleic acid

tion whether irradiation induces qualitative as well as quantitative changes in nucleic acids. Samples of DNA and PNA were isolated from livers of treated mice 18 and 48 hours after irradiation, as well as from control mice. These samples were analyzed for their content of purines and pyrimidines, to detect possible changes resulting from the irradiation.

The methods of irradiation and the strain of mice were the same as those employed in the earlier experiments; twelve mice were used in each group. The liver nucleic acids were isolated by means of a fractionation procedure based on the differential lability of the two acids toward alkaline hydrolysis. The livers were pooled in groups of four to provide sufficient material.

The nucleic acid samples were desalted, partially dried, and hydrolyzed in 70 per cent perchloric acid for 1 hour at 100° C. The nitrogenous bases present in the hydrolyzates were determined by quantitative paper chromatography. dimensional chromatograms were run, using isopropanol-hydrochloric acid as the solvent, and the spots were detected by examination of the dried chromatograms under ultraviolet light. Individual spots were marked and cut out, the bases eluted with o.1 N HCl, and quantitative determinations carried out by means of ultraviolet spectroscopy of the eluates. Eight chromatograms were run of each nucleic acid sample.

Table 7 presents the results of the chromatograms for the various samples. No differences are detectable, and so it must be concluded that, within the limits of the experimental test, no change in the composition of the nucleic acids of mouse liver results from whole-body exposure to 600 r of hard X-rays.

Properties of Mitochondria

Continuing the work reported in Year Book No. 50 on the nature and function of cytoplasmic particles, we again emphasized the study of mitochondria. It was shown in last year's report that the mitochondrial fraction of rat liver, isolated by differential centrifugation from 0.88 M sucrose homogenates, consists of several

particle types differing in chemical properties, and not merely one particle type as had been supposed by previous workers. This study has now been extended to questions of the function and origin, as well as further details of the chemical nature, of these particles.

TABLE 7

Composition of nucleic acids isolated from livers of control and irradiated mice

(Results are expressed as mole fractions of each base. The purine-to-pyrimidine ratios for the pooled data are 1.15 for DNA and 1.17 for PNA.)

	Control	Hours after irradiation		
		18	48	
DNA:				
Guanine	0.22	0.24	0.25	
Adenine	0.30	0.30	0.29	
Cytosine	0.22	0.20	0.22	
Thymine	0.26	0.25	0.25	
PNA:				
Guanine	0.34	0.34	0.33	
Adenine	0.21	0.20	0.20	
Cytosine	0.16	0.17	0.18	
Uracil	0.30	0.28	0.29	

The original mitochondrial preparation (MT_0) , similar to that isolated by other workers, was further fractionated into four parts: heavy particles (MT_1) , which sediment at 3000 × gravity for 20 minutes; light particles (MT_2) , sedimenting at 6000 × g for 20 minutes; still lighter particles (MT_3) , which sediment at 25,000 × g for 20 minutes; and the lightest particles (MT_4) , which are barely sedimentable at 25,000 × g for 20 minutes. These fractionations were carried out with 0.88 M sucrose solution as the suspending medium.

Studies of the distribution of the enzyme D-amino acid oxidase have yielded information of some interest. Results of a typical experiment are shown in table 8,

which includes the phosphorus-to-nitrogen ratios of the fractions, to permit comparison with last year's results. From the distribution of this enzyme, which apparently is concentrated in particles of intermediate weight, it is evident that the various mitochondrial types differ in function as well as in structure. Whether the enzyme occurs at all in the heaviest and lightest particles cannot be decided, since the activity

TABLE 8

DISTRIBUTION OF D-AMINO ACID OXIDASE AMONG FRACTIONS OF MITOCHONDRIA

(Enzyme activity is expressed as µM of pyruvate formed per mg N of fraction per hour.)

Fraction		Per cent MT ₀	
MT ₀	0.95	100	78
$\mathrm{MT}_1.\ldots.$	0.35	37	72
MT_2	1.05	110	73
MT_3	2.27	250	86
MT ₄	0.71	74	118

found in the MT₁ fraction, for example, may be wholly, or only partly, due to the presence of contaminating particles of smaller size.

The distribution pattern of D-amino acid oxidase lends excellent support to the conclusion reached last year, on the basis of studies of absorption spectra, that at least three types of mitochondria exist.

More extensive data are now available on the phosphorus-containing compounds of rat-liver mitochondria. Table 9 presents the results of a phosphorus partition based on the procedure of Schneider. The high protein-phosphorus content of the particles is striking. It can be calculated from these data and from estimates of the total quantity of mitochondria present in liver cells that about two-thirds of the total protein phosphorus of the liver cell is in mitochondria. This result is striking, in view of the very high metabolic activity of such phosphorus.

In contrast with the protein phosphorus, whose distribution seems to be independent of particle size, the content of acid-soluble phosphorus shows a marked relation to particle size. It is difficult to draw any significant conclusions from this observation, however, owing to the large number and diversity of substances making

TABLE 9

DISTRIBUTION OF PHOSPHORUS-CONTAINING
COMPONENTS AMONG FRACTIONS OF
MITOCHONDRIA

	Phosphorus component (μg P/mg N of fraction)						
FRACTION	Total	tal Pro- "Acid-Nucleic tal tein solu- acid ble" acid		Nucleic acid	Lipid		
Whole homogenate.	112	2.3	46	24.8	39		
$\mathrm{MT}_0.\ldots\ldots\ldots$	74	5.2	26	6.2	37		
MT_1,\dots,\dots	52	5.0	15	2.6	30		
MT_2	62	6.0	16	3.9	36		
$\mathrm{MT}_3.\ldots$	74	5.4	28	7.6	34		
MT_4	90	6.1	32	11.2	41		

up this group. Attempts are under way to break it down further, in the hope of obtaining new information about the metabolic activities of mitochondria.

The distribution of nucleic acids is similar to that of the acid-soluble compounds, whereas the phospholipids show the distribution pattern of protein phosphorus, content being independent of particle size. This difference in the pattern of occurrence of phospholipids and nucleic acid is curious in view of the much higher concentration of both that has been reported as occurring in the microsomes (submicroscopic particles). One might suppose that light mitochondria would tend to be similar to microsomes in all respects.

The phospholipid results certainly suggest that very few microsomes are present

in these preparations; any significant contamination by microsomes should be accompanied by a rise in the phospholipid content of the lighter particles. From the results of repeated experiments it appears that the MT₄ fraction may have about 20 per cent more phospholipid than the other fractions. Assuming that this surplus is entirely due to microsomes, it would represent the equivalent of a small percentage of contamination on a nitrogen basis.

A study of the ultraviolet absorption spectra of the cytoplasmic particles has revealed that an unusually strong proteinnucleic acid bond appears to exist there. Figure 8 shows the absorption spectra of the various fractions obtained by dissolving the particles in o.oo1 M KOH. (If the concentration of KOH is raised to o.or M or higher, a typical shift toward longer wave lengths occurs in the spectrum; this is caused by the change in protein absorption occurring in alkaline solution.) All attempts to make an extract of the particles that would give a typical nucleic acid spectrum have failed. Standard methods for decomposing nucleoproteins have no effect on mitochondria. Extraction with boiling trichloroacetic acid; extraction with cold perchloric acid; and incubation in alkali followed by precipitation of the protein using hydrochloric acid and trichloroacetic acid, or adjustment to pH 5 with acetic acid, have all failed to yield an extract whose ultraviolet spectrum differed

significantly from that of the intact particles dissolved in o.oor M KOH. The concept of an unusually strong nucleic acid-protein linkage in mitochondria is

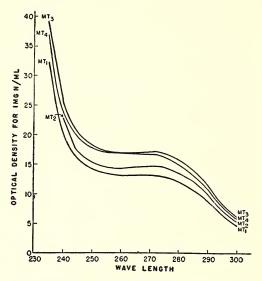


Fig. 8. Ultraviolet absorption spectra of mitochondrial fractions.

supported by elementary analyses of the hot trichloroacetic acid extracts obtained in the course of the phosphorus partition. The nitrogen content of such extracts is several times greater than that which would be predicted from their phosphorus content. Similar extracts of whole homogenate give a nitrogen-to-phosphorus ratio indicating that they contain only nucleic acids—not nucleoprotein, as is the case with the mitochondria.

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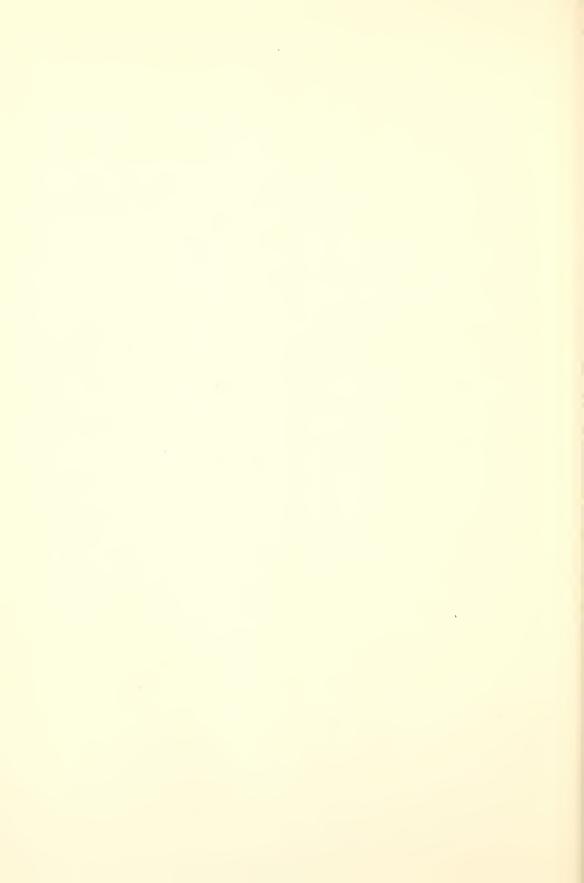
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DEPARTMENT OF ARCHAEOLOGY

Cambridge, Massachusetts

H. E. D. POLLOCK, Director

In the previous annual report we noted the beginning of a new program of researches. It will be recalled that the locus of our studies, in time and space, was the later stages of the preconquest history of Yucatan. Of the several approaches to be employed, archaeological field work was the central one. It was further noted that the ruins of Mayapan would be the scene of our initial efforts.

During the past season these plans were rather closely followed. Most of the staff of the Department were stationed at our base camp at Telchaquillo, adjacent to the Mayapan ruins. In this first period of fullscale operation it was felt that excavation should progress with caution, and that our procedure should be one of intensive archaeological survey using the topographic map of the site that had been completed the previous year as a means to that end. A broad picture of the problems that face us and of the critical areas for investigation has emerged from this study. It will come as no surprise that this exploratory work has raised more questions than it has solved. In the succeeding paragraphs we shall touch upon the wider aspects of our findings and point to some of the problems.

Reconstructions of Maya history have, of course, changed over the years. Such shifting and sifting and rearrangement of ideas is of the very essence of research. It is further true that at no particular time in the course of our studies will there be unanimity of opinion in these matters. At the present stage of thought, nevertheless, few people will differ with the broad chronological and cultural division of the history of the Maya peoples into the pre-

Classic, Classic, and post-Classic Periods, although the divisions will be differently named by different students. There will also be general agreement that we are dealing with a span of time that at least covers the Christian Era up to the European conquest of the New World and that may extend an indeterminate distance into times before Christ. There will be wide differences of opinion as to subdivisions of the larger periods, as to association of cultural periods in different geographical areas, and as to any precise chronology. Concerning no period probably is there greater diversity in our ideas than concerning what we have termed the post-Classic. This was the era that followed the apparent breakdown of the early system of exact dating in the Maya calendar. It is the period with which our present studies are directly concerned.

At this point it seems well to outline, as basis for discussion, a widely accepted reconstruction of Yucatan history during post-Classic times. In our previous report (Year Book No. 50, p. 223) we referred to a Period of Foreign Domination, a Period of Maya Resurgence, and a Period of Disintegration. These three divisions, in the order given, constitute the post-Classic Period in Yucatan. Although he uses a different terminology, the scheme in general follows the ideas of J. E. S. Thompson (American Antiquity, vol. 11, 1945, pp. 2-24). His chronology, in round figures, is Foreign Domination A.D. 1000-1200, Maya Resurgence A.D. 1200-1450, Disintegration A.D. 1450-1540. The Period of Foreign Domination represents the time when invaders of Toltec culture from highland Mexico presumably held sway over Yucatan, their principal base being Chichen Itza. The Period of Maya Resurgence was that during which older, native ideas were supposedly reasserting themselves over the foreign practices of the Toltec, Mayapan reputedly being the governmental center of the northern part of the peninsula. The Period of Disintegration followed the overthrow of Mayapan, involving the end of central authority and the breakup into numerous warring states. This last period was in a sense an extension of the preceding one in that the resurgence of Maya ideas and customs is presumed to have continued. Theoretically, principal settlements of the time should have been the capitals, several of which are known today, of the various states, but up to the present the remains of this particular period have not been identified at those sites or elsewhere in Yucatan (see Year Book No. 50, pp. 227, 232-236).

We may now turn to our findings of the past season and focus these findings against the background sketched above.

It is already clear from our relatively small sampling of the pottery of Mayapan that people lived in or around this site from pre-Classic times, through the Classic Period, and into the post-Classic at least through the Period of Maya Resurgence; as was noted above, the final Period of Disintegration remains archaeologically unknown. This evidence of a long span of occupation agrees with Brainerd's findings of a decade ago (Year Book No. 41, 1941-1942, pp. 254-255). Pottery of pre-Classic times occurs as little more than a trace, and pottery that might date from the early part of the Classic Period is relatively rare. No remains of buildings recognizable as belonging to either epoch have been found.

It was during the later phases of the Classic Period that Mayapan seems first to have become a center of any importance. In Yucatan this period is known as that of the Puuc, taking its name from the hill area along the border of the States of Yucatan and Campeche, in which the greatest concentration of remains of the time is found. It was a time of great vitality, and architectural remains of characteristic Puuc style occur in huge numbers in that area and sporadically throughout the northwest part of the peninsula. The period might be dated from A.D. 700 to A.D. 1000, although there is little basis for any exactitude in chronology. The era comes to a close with the supposed abandonment of the hill area and the end of the Puuc architectural style. At Mayapan we find Puuc Period pottery, little in proportion to that of the later, great period of the site, but nevertheless in appreciable amounts. Possibly more striking are the widespread building remains. Not a single standing structure of the period has so far been found, but building stones, both carved and plain, fashioned in the typical Puuc manner occur all over the ruins, lying individually and in groups, and re-used in later construction. Fortunately, in many cases the stone work of the period is unmistakable and can be identified with little hesitation. There is also the possibility that some of the stelae and other pieces of sculpture at Mayapan may be of this time.

We find the opening phase of the post-Classic Period, what has been termed the Period of Foreign Domination—the Toltec Period at Chichen Itza—virtually unrepresented, or at best but slightly represented, at Mayapan. This is surprising, not only because historical sources suggest that the city was then of some consequence, but because the site so clearly had been occupied in the immediately previous period. Nevertheless, no example of architecture or of sculpture can surely be assigned to this epoch. A few such remains are reminiscent

of Chichen Itza, the type site of the period, but even in these cases there is sufficient difference in execution to question their being products of the time. Unfortunately, we are unable to present a clear picture of the evidence regarding pottery. Two imported types that presumably are indicators of this period occur, or at least are recognizable, in trace amounts. They are conspicuous more by their absence than by their presence. No ceramic stratigraphy has thus far developed at Mayapan, and in our present state of knowledge we are unable to sort out the local Yucatan pottery that might be assigned to the period.

Our next division in time, the Period of Maya Resurgence, seems quite clearly to include the rise to power and greatness of Mayapan. Most, if not all, of the standing or partly standing architectural remains, much of the sculpture, and the vast preponderance of the pottery belong to this time. There are faint indications at Mayapan that this was not an altogether static period, that culture was undergoing changes reflected in the remains, and that we may be able to divide the period into phases. Little, however, can be said at present on this subject.

Let us summarize in a few sentences the picture we have sketched of our preliminary findings, and then turn our attention to a broader scene. The peoples of the pre-Classic and Early Classic Periods left but a trace of their handiwork at Mayapan. The Puuc culture, presumably of Late Classic Period, is clearly represented in architecture and pottery. This seems to have been the time when Mayapan first became a center of any importance. During the first stage of the succeeding post-Classic Period, the Period of Foreign Domination in Yucatan, Mayapan appears to have been of little or no importance; indeed, there is even the suggestion of a break in cultural continuity at this time. The Period of Maya Resurgence, possibly divisible into phases, witnessed the greatness of Mayapan. This is attested by the building remains and by the wealth of pottery recovered. It has been noted earlier that no archaeological remains, at Mayapan or elsewhere in Yucatan, have surely been recognized as belonging to the final Period of Disintegration.

Turning to the wider scene, the history of northern Yucatan, we shall confine ourselves primarily to the later stages that together form the post-Classic Period, the times with which our studies are directly concerned. We have already described briefly the immediately preceding, presumably late Classic, Puuc Period, and pointed out that, although the home area of this culture contains by far the greatest concentration of remains of the period, traces of the characteristic architectural style occur well to the north and in general may be said to be spread over the northwest quadrant of the peninsula. In the region north of the hills, there also occur architectural forms strikingly different from those of the Puuc. In certain instances these underlie the Puuc remains. There is some reason to believe on stylistic grounds that these ruins are of the Early Classic Period, but their date is by no means certain, and there is the possibility that they are Late Classic, conceivably contemporaneous, or in part contemporaneous, with some of the Puuc ruins in the hill region.

The eastern extension of the Puuc culture is not well known. Architectural forms that vary from a general similarity to those of the Puuc to virtual replication occur at Chichen Itza. This is the approximate eastern boundary of this architecture as now known, but the characteristic pottery has been found farther east. The northeastern part of the Yucatan Peninsula is not well explored, and there is ample

room for future discovery. Should present indications that Puuc styles of architecture do not occur in the area be confirmed, we hope to learn what building forms were being used at this time, and what are the stylistic affiliations to other areas.

It will be remembered that the early post-Classic was the Period of Foreign Domination, according to the scheme we have been following; it has also been called the Mexican Period and the Toltec Period. It was at this time that Chichen Itza seems to have reached its apogee, the greatest structures there being assigned to this epoch. Although the concept of a foreign element in Yucatan arises in part from documentary and historical sources, the primary evidence pertaining to the period under discussion consists of the architectural and sculptural remains at Chichen Itza. These exotic forms suggest ideas, religious beliefs, and social organization that are not Maya. The amazing similarities to the art and architecture of Tula, Hidalgo, moreover, leave little doubt as to the identity of the invading culture.

The archaeological evidence is bolstered by documentary and historical references associating Chichen Itza with what seem to have been non-native peoples, although the time of the association is open to doubt. There remains the question of how thoroughly the foreign culture penetrated the social fabric of the city and its environs, and whether the non-Maya elements affected only the priestly and ruling group or were taken over by the commoners as well; but the presence of a foreign culture at Chichen Itza seems indisputable.

In northern Yucatan, we find no such evidence. On the contrary, we are presented with a situation closely parallel to that at Mayapan. Outside the immediate area of Chichen Itza, with the exception of a few traces—and those of questionable identification—there is not a single ruin,

so far as we know, that exhibits architectural or sculptural remains that can be identified as like those of the type site or as being of this period. We do not know at present what is the pattern of distribution of pottery. As a result of his work of some years ago, Dr. Brainerd seems to think that the typical pottery of Chichen Itza of this period is by no means confined to that site (Year Book No. 41, pp. 253–257).

We have, then, a most puzzling picture of a single great city that appears to have exerted little or no influence on the arts and handicrafts, with the possible exception of pottery, outside its immediate neighborhood. The situation becomes no less puzzling when we consider the vitality of the Chichen Itza culture of this time. Taking our evidence at face value, we are forced to picture a great, thriving city existing in a sort of cultural vacuum. Whether this be interpreted as meaning that human effort from all parts of Yucatan went into the building of Chichen Itza, to the exclusion of local works, or that the dominance of that city over the peninsula was so great that nowhere else were people allowed or able to express themselves in the new architectural forms, we are presented with a picture that lacks reality. The presence elsewhere in Yucatan of pottery of a sort made at Chichen Itza at this time, moreover, would not alter the situation. This fundamental and simple handicraft presumably would continue no matter what was taking place in the higher expressions of art and architecture.

Here is an interesting problem. The nature of the question and its answer hinge upon our chronological framework and how we interpret events during the Period of Foreign Domination. How widespread in Yucatan, and of what effect upon the social structure, was foreign influence? How rapidly did the conquest, if in fact

there was a conquest, proceed? Are we correct in assigning the foreign penetration to a period following that of the Puuc? The greatness of Chichen Itza, expressed in its architectural remains, and certain historical references to the dominant role of that city at a certain period in history have led many people to believe that a great part of the peninsula was under the sway of the foreigner. Yet we have just noted that the archaeological remains, as known today, do not bear this out, unless we think of the invasion as a military conquest of such intensity that virtually all important effort in architecture and sculpture ceased outside Chichen Itza. It should be remembered that we are not dealing with a period of a decade or of a generation, but with one of approximately 200 years.

There are, of course, a number of ways in which we can modify our ideas in respect to the political and cultural events of the time. We might suppose that the conquest of Yucatan was relatively benign, a supposition that has little basis in the light of the militaristic character of the Chichen Itza sculptures. We might assume that the invader exerted political control over the country but had little effect on the native culture. This also is difficult to believe because of the warlike character of the foreigner and the length of time involved. In the case of either of the above suppositions, we are plagued with the riddle of no material remains, except possibly of pottery.

A position that seems more tenable is to assume that the invader and his culture were confined to the Chichen Itza area or possibly the eastern part of the peninsula. This would not be unlikely if the foreign elements entered Yucatan when the Puuc culture was still vital. In any event, the older forms of art and architecture might have continued in use in the west. We then become involved, however,

in further questions: Precisely what, for example, in time and culture, does the Puuc Period represent? When did it terminate? When was the Puuc area abandoned? Did the characteristic architectural style continue in use elsewhere, particularly in the north? These questions—and many more could be formulated—are subject to answer only by further exploration, excavation, and study of existing data. Our purpose here is simply to note that the apparent absence at Mayapan of the remains of an important and critical period has led us to look further and, in so doing, to question the time and the nature of certain events in the history of Yucatan.

The next division in the chronological framework we have been using, the Period of Maya Resurgence, presumably saw the rise to power of Mayapan. It is often referred to as the Mayapan Period, that city being the type site. Elsewhere in Yucatan, notably along the eastern coast, there occur architectural remains that stylistically and in method of construction are very similar to those of Mayapan. Certain presumably late constructions at Chichen Itza may well belong to this period, particularly to its earlier part. What little is known of the pottery seems to confirm the placing of the East Coast and late Chichen Itza remains in this general period. Other occurrences in Yucatan of constructions and pottery of Mayapan type are sporadic and often questionable as to identification.

Although one might expect more numerous traces of the culture of these times, the lack of evidence does not appear comparable to that concerning the preceding period. Structural design of buildings was inferior to that of earlier times; disintegration undoubtedly proceeded rapidly, and little was left comparable to the earlier distinctive stone cutting and typical architectural elements. There is also the suggestion that sizable ceremonial structures

were not being built in any numbers, and small constructions, domestic or religious, are not readily spotted in the course of exploration. Possibly of even more significance in this connection is the fact that we are dealing with a period of cultural decadence. It would not be unexpected that in such times the effort that went into religious or public monuments might largely be confined to the central seat of authority or at most include a few of the larger provincial centers.

There has been a tendency among Mayanists in recent years to divide what we have called the Period of Maya Resurgence into two subperiods, the division falling around the middle of the fourteenth century. It has been mentioned that at Mayapan there are faint indications that the remains of the principal occupation of the site may reflect two cultural phases. Such a division would seem better to suit possible ceramic and architectural relationships between Chichen Itza and Mayapan. Our evidence at the moment is much too fragmentary to draw conclusions, but it is an idea with sufficient basis to merit attention.

A matter of equal importance with chronology is the character of the culture at any time. The two periods we have primarily been discussing have been labeled Foreign Domination and Maya Resurgence. Though these terms are in a sense no more than convenient tags, they carry cultural or political connotations and deserve examination in that light. The Period of Foreign Domination has been discussed, and the appropriateness of the term has been brought into question. The designation of the second period, Maya Resurgence, is a sequel to the first, the implication being that foreign forms and practices gave way to older, native ideas. This has also been called the Period of Mexican Absorption, with much the same implication. Some students have preferred to label both periods simply as Mexican, not recognizing, at least in designation, the idea of a return to native Maya traditions.

The concept of a Period of Maya Resurgence or Mexican Absorption is based on historical, ethnological, and linguistic evidence at the time of the Spanish conquest and on the absence of art, particularly sculpture, like that which flourished at Chichen Itza in the preceding period. There is also the evidence of presumably late art at Chichen Itza, and possibly in the East Coast ruins, showing more Maya elements than the Toltec art of that city. On the other hand, the great preponderance of building remains assigned to this late period are not of Maya, but of so-called Mexican, style. Though the architecture differs from that of Chichen Itza of the preceding period, the style could well be a degeneration of those forms and certainly is more foreign than Maya. Indeed, in respect of construction this late architecture is farther from the Maya than are the earlier Chichen Itza forms. The question arises, then, Are we correct in thinking of this period as a time of general resurgence of Maya culture throughout Yucatan?

The answer is clearly affected by our ideas concerning the preceding period. If the foreign culture was not generally accepted in Yucatan but was confined to the Chichen Itza region, we can hardly speak of a Maya resurgence except as applied to that city. Even if the invaders controlled a larger area, there is considerable question how much of their culture was taken over by the Maya, and whether the civilization of Yucatan was more foreign then than in later times. Indeed, one can take the position that foreign cultural elements were more common in the later period than in the earlier. These questions are not now answerable with any certainty. The problem of relative chronology, raised above, is of paramount importance. The conclusion we reach is simply that, taking the hard core of evidence now available, one well can question certain widely accepted beliefs concerning the late preconquest history of Yucatan.

Our idea in the foregoing discussion has not been to sketch the history of those times or to present all arguments and each alternative. The intent has been to indicate on a broad scene some of the problems with which we have to deal and the status of those problems at the beginning of our present program of research. We now may turn to a review of certain staff activities of the past year. Detailed reports by members of the staff will be found following the present section.

The off-season period, July through December, was used by Strömsvik for the construction of our base camp at Telchaquillo. The essential work was accomplished by the first of the year, but throughout the winter and spring Strömsvik was engaged in adding minor conveniences, in adjustment and maintenance of machinery, and in the supply of special equipment for the need of the moment. Experience of a season indicates that the camp is comfortable, convenient, and well adapted to our needs.

In the latter part of November, Strömsvik flew to Honduras to confer with governmental authorities relative to problems concerning the antiquities of that country. He participated in discussions leading to the establishment of an Instituto de Antropología e Historia similar to such bodies in Guatemala and Mexico. Strömsvik has been named Technical Adviser to the Institute, a position involving voluntary services.

It was noted in our previous annual report that the ordering and disposal of large archaeological collections from earlier years had virtually been completed, and that all suitable objects had been put into the hands of the Instituto de Antropología e Historia de México for display in their National Museum or the Merida Museum, or for study purposes. During the past season R. E. Smith completed the assembling of ten collections of sherds, each collection indicating, as far as possible, the known types and range of Yucatan pottery. This work had been started the previous season by R. H. Thompson. Three of the collections were made available to the Institute, one to be sent to the National Museum of Guatemala, the other two for use in the National Museum of Mexico and the Merida Museum. The remaining collections were sent to museums in the United States. With the exception of sherds kept for study purposes, the Department has now turned over to the Mexican Government all archaeological material collected prior to 1951.

During the 1951 field season the work of Ruppert and A. L. Smith on the house platforms at Mayapan (Year Book No. 50, pp. 15, 231) developed the fact that these dwellings were strikingly similar to the native houses described by the sixteenthcentury historian Landa. Further confirmation of such historical record occurred this year. Landa observes that the people buried their dead in, or at the rear of, their houses. Excavations of Smith and Ruppert have uncovered several cists or chambers below the floors of dwellings. Interestingly enough, these "graves" were built at the time of the construction of the house, apparently in anticipation of the death and burial of the occupants. That the "graves" were not always used, however, is shown by the fact that in only three of seven examples uncovered were human remains found.

Landa also mentions that "the nobles and persons of high esteem" were cremated rather than interred. Although little work has so far been done at Mayapan in connection with ceremonial or public buildings, Shook discovered below the floor of what apparently was a temple a repository containing the charred bones of adults, children, and animals. This evidence, and that developed by Ruppert and Smith, is suggestive, but at present our knowledge of the burial customs of the inhabitants of Mayapan is fragmentary, much too meager to warrant any general conclusions.

The mapping operations of Jones in 1950 and the archaeological survey of Ruppert and Smith in 1951 disclosed the existence at Mayapan of numerous rough stone walls that may have marked the boundaries of properties surrounding dwellings (Year Book No. 50, pp. 226, 232). W. R. Bullard, Ir., a graduate student in anthropology at Harvard University, working with this Department, began a study of these walls during the past season. Preliminary results indicate that the walls occur in residential but not in public areas, and that they may indeed form the boundaries of house lots. Information relative to the pattern of these walls, of passages between properties, and of streets or lanes is beginning to emerge. Fulfillment of the underlying objective of this research, an understanding of city planning, land tenure, density of population, and similar problems, must await further studies.

Proskouriakoff has undertaken the recording and study of the not inconsiderable body of sculptural material found at Mayapan. This has proved to be a disheartening task because of the lamentable condition of most of the sculpture so far discovered. An interesting possibility is that stone figures found in some numbers can be identified as the "idols" reputedly introduced into Yucatan in late times as part of the foreign culture mentioned earlier in this report. The practice of "idol-

atry" apparently was deeply resented by the older Maya. Proskouriakoff has also begun the study of the artifacts—implements, weapons, minor carvings, and the like—of Yucatan.

The existence of another walled city in Yucatan, reported a century ago by an anonymous antiquarian of that country but never visited, so far as we know, by archaeologists, was confirmed by the explorations of Strömsvik and Pollock. Walled cities, few of which are known in the lowland Maya area, are important because of certain implications as to the civilization of the builders. The wall at Chacchob, as the site is known, is very similar to that of Mayapan, but the architectural remains and a small sample of pottery seem to antedate the great period of the larger city. This is surprising, as walled cities have been thought to be phenomena of later times. Further exploration and testing at Chacchob are needed to ascertain the period of occupation of the site.

Though Shepard gave a great part of her time to the completion of a book on ceramics for the use of archaeologists, she also was able to continue her researches on the technology of Yucatan pottery. This work has been broadened in scope by the examination of collections of pottery from areas outside the peninsula that may have exported pottery to Yucatan, and by studies of present-day native pottery and pottery methods in Yucatan and Campeche. The possibility that there may be relatively few usable sources of clay in northern Yucatan suggests that the region is particularly suitable to technological research. Certain peculiarities of Yucatan ceramics, such as the high proportion of macroscopically indistinguishable wares and the frequent difficulty of obtaining adequate stratigraphy, combined with indications of distinctive but little-understood regional developments, make the technological studies of special value to our present work.

The large undertaking of compiling a Maya hieroglyphic dictionary, described in last year's annual report (Year Book No. 50, p. 244), was continued by J. E. S. Thompson. Work on texts at Yaxchilan, a large site on the Usumacinta River, brought to light interesting material on the brief adoption by that city of the socalled Puuc style of recording dates, a method known to occur elsewhere only in Campeche and Yucatan. During the summer of 1951 Thompson visited Yucatan, Tabasco, and the famous ruins of Palenque in Chiapas. At the latter site he recorded a group of magnificent hieroglyphs in stucco, discovered many years ago in Temple 18 but never previously photographed. Thompson is now engaged in writing a popular book on Maya civiliza-

Continuing his researches on the historical geography of Yucatan at the time of the conquest, Roys, operating under a post-retirement grant, spent two months in the field. Of particular note is his identification of what probably was the site of Sinsimato, a large and important town in northeastern Yucatan at the time of Montejo's first expedition early in the sixteenth century.

Studies by Morris on the prehistory of the southwestern United States have resulted in the completion of a definitive report on an important group of Basket Maker II sites near Durango, Colorado. A late development in this work was the discovery that certain wood samples collected by him, and dated by dendrochronological methods, extend the known range of prehistory in that area 171 years farther into the past, namely, to the year A.D. 46. There is the excellent possibility, moreover, that

some of Morris' samples will eventually be found to date at least 100 years earlier, well before the birth of Christ.

During the year Joseph A. Hester, Jr., has been appointed a Carnegie Institution Fellow for studies of geographical conditions, with particular reference to agricultural potentials, that may have affected the economy of the ancient Maya. A report on his work, as well as a report by R. H. Thompson, whose Fellowship has now terminated, will be found in succeeding sections.

The Mesa Redonda sessions of the Sociedad Mexicana de Antropología, held in Jalapa, Veracruz, the latter part of July 1951, were attended by several members of the staff. Papers were presented by Proskouriakoff, "Scroll patterns (entrelaces) of Veracruz," and by J. E. S. Thompson, "Relaciones entre Veracruz y la región Maya." Shook presented information covering the distribution and chronology of vokes and thin stone heads in areas southeast of the Isthmus of Tehuantepec; R. E. Smith discussed certain similarities between the pottery of Veracruz and that of the Maya area. Shepard presented an exhibit showing the effect of different firing temperatures on different clays, a matter of importance in connection with the classification of pottery.

MAYAPAN, YUCATAN

A. L. SMITH AND KARL RUPPERT

During the 1952 field season, January 17 to May 1, A. L. Smith and Ruppert continued their surface examination of the ruins of Mayapan (see Year Book No. 50, pp. 230–232). Squares F, G, H, I, O, P, and R were covered this year (Jones, 1951, C. I. W. Notes, no. 106, map). All structures outside the wall within the limits of the map, in squares so far completed, were

examined and numbered. In the course of the season's work 1165 buildings were recorded. These, with the 1143 examined in 1951, leave approximately one-third of the site yet to be investigated.

Every structure seen was given a number, but only those which are of the dwelling type or those associated with such groups were drawn to scale. The structures examined fall into the general categories established during the 1951 season. Some variations were found, but no new types. The recurrence of types recorded during the first season, however, their association with one another in groups, and the distribution of these types in various parts of the city are beginning to show a definite pattern. This pattern will undoubtedly become clearer when the survey is completed and a definitive study has been made.

The location of metates, when associated with structures, was recorded. These utensils usually occur on platforms or terraces and are often re-used in construction. The occurrence of carved stones and Puuc-type building stones was also noted. The latter are found in some quantity re-used in many of the buildings, their presence predominating in the more elaborate structures.

Though the major part of the season was devoted to the examination of the structures mentioned above, several weeks were spent excavating below the floors and benches of house platforms and associated terraces. Bishop Landa, in his Relación de las cosas de Yucatan, mentions that the dead were buried in their houses or at the back of them (Tozzer, 1941, Papers Peabody Mus. Harvard Univ., vol. 18, p. 130). With this in mind, several types of construction were selected for excavation: long, low platforms and houses with two and three benches. Twenty-two pits were sunk in benches, passageways, back rooms, and terraces in front of buildings. Masonry-lined cists were found under five

benches and two platforms. These varied greatly in construction, from a nicely plastered rectangular chamber with corbeled vault to rough, unplastered compartments filled with rubble. Three of these cists had jambs in one end, indicating an entrance below floor level.

In all cases the cists were built at the same time as the structure in which they were contained, apparently in preparation for the interment of the occupants of the house. Only in three of them, however, were human remains present. Little grave furniture was recovered with the burials. In one tomb, where there were two skeletons, two pottery figurines, male and female, were found. These were possibly the idols of the individuals buried, in accordance with a custom mentioned by Landa (*ibid.*).

Until more excavation has been carried on in house platforms, very little can be said about burial customs of common people at Mayapan.

In all instances the latest-type pottery recovered from any dig was of the Mayapan period. It is interesting to note, however, that in one case (Structure J-49-a) the Puuc type of pottery predominated (see p. 253).

A rewarding find was a midden under the east bench of Structure J-50-a. From the pit below this bench a quantity of artifacts was recovered in addition to a large sherd collection. Among the artifacts were worked shell and bone (the latter in the form of awls and needles), obsidian blades, human teeth perforated for suspension, flint implements, a stone metate leg, and miscellaneous animal and bird bones.

BOUNDARY WALLS OF MAYAPAN

WILLIAM R. BULLARD, JR.

Work of the previous season at Mayapan disclosed the existence of numerous fieldstone walls running everywhere among the ruins (Year Book No. 50, pp. 226, 232). Whereas some walls were obviously of postconquest construction, others were quite clearly contemporary with the occupation of the site. Because many of the latter appeared to surround house structures, it was thought they might delimit property boundaries. During the season of 1952, all the walls in certain representative portions of the site were followed out and plotted on the map. It was hoped that such a study might reveal with certainty the function of the walls and cast light upon land tenure, city organization, and similar problems. Although stone boundary fences around the houses are a regular feature of modern Yucatan villages, they have not previously been reported from a preconquest Maya site.

The walls were completely plotted in roughly one square kilometer of the site, comprising Squares D and K in the north central part and Squares Z, AA, BB, DD, and EE in the southwestern part (Jones, 1951, C. I. W. Notes, no. 106, map). In addition, other areas of Mayapan were reconnoitered in order to estimate the value of additional work.

The entire site is covered by what seems a maze of stone walls, ranging in age from the occupation of the city to the present day. For many years Mayapan was the site of a Spanish cattle ranch, and stone corrals and enclosures were built throughout the central part of the ruins. Moreover, probably ever since the conquest there has been milpa cultivation within Mayapan, as there is today. Finding plentiful stone at hand in the ruined buildings and walls, the Maya farmers often erect stone fences around their fields, not hesitating to repair and re-use older walls when convenient. Centuries of such destruction and rebuilding have confused to a large extent the ancient wall patterns. Relatively few ancient walls are complete,

and in many instances only short sections could be followed. Even fragmentary walls could usually be related to house structures, but the complete pattern over a large area was all too often obscured. In this respect, the walls plotted in Squares AA and BB proved most satisfactory, since disturbance in those sections has been relatively slight.

Another perplexing problem was that of distinguishing ancient from postconquest walls. Technologically there are few reliable criteria that can be applied to tell a postconquest wall from a re-used and rebuilt ancient wall, or an ancient wall from an old and much damaged postconquest wall. Thus, although many, indeed the majority, could be identified without difficulty, many others had to be marked as questionable. All walls, both ancient and modern, were plotted, by use of different symbols, in the hope that the pattern followed by a questionable wall would give a clue to its origin.

In construction, the ancient walls are essentially alignments of uncut stones set on edge, either touching or placed a few centimeters apart. The average height is 50–70 cm, but some walls are composed of large slabs up to 1.5 m high. In areas free of postconquest disturbance, the walls often have smaller stones lying near their base, suggesting that the spaces between the slabs were chinked and filled, but the debris was never enough to warrant the belief that the walls rose substantially higher than the stones now standing.

In nearly all instances the walls partly or completely surround individual dwelling-type houses or house groups. Only rarely is more than one house group surrounded by the same wall. Apparently, almost every domestic structure had a boundary wall. In contrast, temples and public buildings such as colonnades do not seem to have been surrounded by these

walls, although further checking is needed to confirm this point.

The walls ordinarily enclose only enough space for a small yard around the structures. There are numerous variations, but most enclosures are of fairly simple plan, consisting of a rough circle, or rectangle with rounded corners. The houses may be completely encircled or the walls may abut against a house or terrace corner so that they are only partly enclosed. In the more densely settled districts, where space is limited, the enclosures tend to become more irregular, and two neighboring house groups may share a common wall.

Gateways, consisting of a gap about a meter wide between two large stones, were found only rarely. The location of many has undoubtedly been obscured by the collapse of wall stones or by postconquest rebuilding. Some enclosures, however, could be entered through larger gaps often formed by the overlap of the two ends of the encircling wall. In a few instances the enclosure may have been reached by passing through the house itself.

There seems little doubt that the walls mark property boundaries around the houses. Evidently each household or group of families had its own fenced yard. This does not, however, necessarily imply that there was private ownership of land in the modern sense. It is quite possible that property was held only by permission of higher authority. Many yards are sufficiently large and on suitable terrain for the raising of fruit trees and small gardens; others include only the tops of the rocky hillocks which were the preferred house sites but which seem unsuited for horticulture.

Nothing was found in the areas plotted resembling a formal arrangement of streets. Passage through the city was evidently accomplished by winding through the gaps and spaces between neighboring boundary walls. Sometimes, particularly where houses are close together, the parallel walls of adjacent enclosures form short, often meandering, lanes averaging about 2 m in width. In one densely settled region in Square AA a particularly complex system of lanes was discovered.

Cenotes are commonly in the areas between house boundary walls and thus accessible to the general populace, as might be expected.

An interesting feature is the small stone circles, averaging 3–5 m in diameter, which are occasionally found near house groups. Some are almost perfectly circular; others are more oval. There is often no gap which might have served as an entrance.

On completion of the detailed examination of the walls in the squares listed above, a reconnaissance was made through other areas of Mayapan. In the eastern part of the site several long lanes were found which, unlike the lanes described above, seem to run independently of boundary walls. The most interesting are in the northern part of Square I, where four lanes converge on a small natural depression with three or four steps leading into it—possibly a blocked cenote entrance. Two of the four lanes are part of a series which could be followed in an almost straight line for nearly 400 m through an area in which houses are rather sparse. Additional work will be necessary to clarify the function of these lanes and their position in the over-all settlement pattern.

As has been mentioned, postconquest disturbance in the areas investigated has been considerable. Although much knowledge concerning the boundary walls has been gained, present information is not clear enough to answer conclusively such specific questions as those regarding the correlation of size of property with size

and elaborateness of the house groups, or the relation of the boundary walls with natural features such as depressions which could have been used for growing fruit trees. Disturbance seems, in general, to be somewhat less in the eastern part of Mayapan, and it seems certain that further work there will produce much additional knowledge.

THE GREAT WALL AND OTHER FEATURES OF MAYAPAN

EDWIN M. SHOOK

Shook arrived in Merida from Guatemala on January 13 and returned to Guatemala on May 18, 1952. The major portion of his time in Yucatan was devoted to researches at Mayapan. These included a detailed investigation of the great wall surrounding the ancient city, the partial excavation of two temples, a brief reconnaissance of the cenotes within the walled enclosure and several outside the wall, and study of Mayapan pottery in collaboration with R. E. Smith.

The great wall of Mayapan, first explored by Ralph T. Patton in 1938 (Year Book No. 37, pp. 141-142) and later by Morris R. Jones (Year Book No. 49, pp. 194-197; No. 50, pp. 229-230, and C. I. W. Notes, no. 106) now appears as a ribbon of stone rubble meandering over 9 linear km through the low, dry, and exceedingly thorny Yucatan bush. In plan it forms a huge, irregular oval with the sharp end pointing slightly north of east. The wall consists of an outer, higher, primary wall and a lower one built against the inner face. This formed a single wall, very thick at the base and stepped at the top, the lower step being a bench, the higher step a parapet.

This wall, representing a vast expenditure of human labor, was built of rough limestone blocks quarried from the out-

crops of bedrock nearest the vicinity of construction. The stone was utilized as it came from the quarry, without further shaping or dressing, and laid without mortar. Parallel lines of large stones, placed either horizontally or vertically, formed the wall's facing and retained the core of dry rubble. Presumably the top of the wall, both the parapet and the bench, was capped with a layer of lime cement, though no evidence of this could be obtained, as no part of the wall's entire length was preserved to its original height. Disturbance by tree roots during the centuries has reduced most of the wall to a low line of stone rubble. A few parts still stand to an average height of 1.5 m; an occasional section where the wall crosses a low area reaches 2 m.

A total of seven major and five minor passages were discovered at intervals in the great wall, and others may have existed formerly. The uncertainty regarding the original number is due to the breaching of the wall in postconquest times for roads and trails crossing the site. These openings vary in width, and it is possible that some of them obliterated ancient minor passages. One major and one minor gate of the twelve identified had been blocked with masonry during the occupation of Mayapan. This presumably left the remaining six major and four minor gates in use simultaneously. During the field season all these were examined, though only four major gates and one minor one were excavated intensively.

The minor gates are straight, narrow passages about 1 m wide, their floors surfaced with lime cement. The major ones are distinguished from these by a considerable thickening of the great wall on each side of the gateway and by a more elaborate plan. This consists of an entrance about 2 m wide through the wall's outer

part, leading to an amplified rectangular space suggesting a portico but still set within the wall. The portico may have one or two columns placed in line with the inside edge of the wall. The placement of these columns suggests that they may have supported timbers for thatch, if the gateway was roofed. The excavation of the lime concrete floors of the porticoes disclosed no debris from masonry vaults or beam-and-mortar roofs.

Careful search was made in a large area around the gates and within the passage-ways for indication of ceremonial activities, guardhouses, arsenals, or warfare. No evidence was found of ceremonial pottery dumps, of guard- or storehouses, or of battles at or in the immediate vicinity of the gates. The pottery and artifacts recovered were exceedingly meager in quantity and widely scattered. On the floor within Gate T, at the southeast extremity of Mayapan, however, were discovered several fragments of a thin gold object and a pellet rattle from a copper bell.

Near the same Gate T and just within the great wall of Mayapan are two structures on the southern side of a large cenote, X-Coton. Both these structures were briefly investigated. The western one consists of a terraced, rectangular substructure about 3 m high with a stairway on the north side leading from the plaza level to a single-room temple above. The latter has thin, low masonry walls, which when built probably stood no more than 50 cm high. The temple walls above this may have been of upright wood poles supporting a thatch roof. In the center of the temple room was a roughly circular shaft extending downward about 1.5 m into the dry rock fill of the substructure. The shaft had been opened in modern times and its contents disturbed and partially removed. We were able to determine from the residue in the shaft that it had been used as a repository for the ashes and charred bones of cremated adults, children, and animals. Mixed with the ashes and bones were fragments of pottery vessels, incensarios, obsidian flake-blades, and flint chips. Some of the pottery vessels once may have contained the cremated remains; but as all were found broken, it is uncertain whether the breaking had been done by the modern looters or by the Mayapan people at the time of burial.

The second structure, located just southeast of Cenote X-Coton and nearer Gate T, was investigated more thoroughly. Here sufficient excavations were carried out to remove the accumulated surface debris from the frontal stairways, the summit terrace, and the temples, and to obtain an accurate plan, section, and elevation of the unit and some evidence of its function. This structure consists of a long, low, rectangular platform supporting twin temples. The temples face northwest, and each has its individual stairway with balustrades leading from the general plaza level to the summit of the common substructure. The twin temples differ only in a few details. Each has a single room with an altar against the center of the back wall. Also, each has low, relatively thin masonry walls built of a few large uncut blocks, many roughly shaped blocks and slabs, and some well cut, faced, and carefully dressed stones. These oddly assorted stones were piled up rather haphazardly, though apparently bedded in a small amount of mud mortar or mud mixed with lime or limestone gravel. This type of masonry appears to be that used in Mayapan most frequently for walls of buildings and facing of substructures, and may be termed "Mayapan block and slab" masonry. The sporadic occurrence of quite obviously reused, well finished stones among the

rougher ones has chronological and cultural implications. These stones are readily distinguished from the others by their form and surface finish. They included wall, vault, and plain or decorated cornice stones, and their original use had been in vaulted buildings whose walls were faced with veneer stones belonging to the Puuc style of Late Classic Maya architecture. Remains in this style point to an older period of occupation at Mayapan, long antedating, we believe, the establishment here of a large, important political center. The people responsible for the later city had lost either the knowledge of architecture possessed by their predecessors, or the desire or the economic ability to equal the quality of the older civic and religious buildings.

It was certain from the shallowness of debris covering their floors that neither of the twin temples had been vaulted. The northeast temple, the larger of the two, contained extremely little debris and probably had been roofed with thatch. Also, because of the wide breadth between jambs, two equally spaced wooden posts must have served as doorway columns.

The smaller, southwest temple may have had a beam-and-mortar roof. Its floor was buried beneath about 50 cm of fallen masonry, including what appeared to be roof material. This temple had undergone at least five alterations, each marked by the addition of a new lime concrete floor and minor revisions of the masonry altar against the back wall. All these floors and the successive altars, normally white, were severely burned to a grayish black. The frequent renovations of the temple's interior were required possibly by numerous ceremonial fires which smoked the walls and heavily stained the floors. Many fragments of pottery incense burners were found on the surface of the top floor and on the frontal terrace outside. Quantities of the same material also lay on the floor of the northeast temple, on the stairways, and along the base of the substructure, but only on the floors of the southwest temple was there evidence of burning. Over 50 per cent of all pottery recovered from the excavation of these twin temples consisted of fragments of incensarios.

A miniature shrine with a narrow doorway was uncovered, centrally placed, at the base of the northeast stairway. Its thin, low stone walls were constructed partly on the plaza floor and partly overlapping the lower steps. No comparable shrine was found associated with the southwest stairway. Instead, in front and on the plaza level, there were three masonry altars placed on the central axis of the temple. The altar farthest from the stairway was the largest of the three. Though considerably damaged in its exposed position, it appeared to be about 4 m square and 50 cm high. Against the altar's edge, facing the temple, a plain, crudely cut stela still stands in its original position. The middle altar, also square and of about the same height, measured 2 m on a side. The one closest to the temple stairway was originally a single circular stone, plastered over and painted bluish green. This altar underwent several remodelings and in its final stage was square. The evidence of much ceremonial activity in the southwest temple, plus the alignment of three altars and a stela in front of its stairway, suggests that this was the more important of the twin temples.

Some attention was given to the cenotes within and immediately outside the walled area of Mayapan. Altogether, 22 cenotes were studied briefly for the purpose of determining which the Maya had used for water supply and which had associated

constructions or contained evidence of ceremonial rites.

During this investigation it was learned from local informants that each year, generally in the month of August, the people of Telchaquillo hold a rain ceremony at Cenote Itzmal Ch'en, the most easterly one inside the Mayapan wall. Here, 3 km from their homes, secluded beneath several fine shade trees overlooking the beautiful cenote and surrounded by ruined temples and houses of their forefathers, the living Maya still call on their gods of the forest, the winds, the milpas, and especially the rain to aid the young corn to grow strong and produce abundantly. All females and children are excluded from this solemn affair. Each man contributes an equitable share of the money, alcoholic beverages, candles, turkeys, chickens, corn, and other items demanded by the professional shaman, who is hired from the town of Tecoh to conduct the ceremony. The ritual begins at sunrise on the appointed day and continues without interruption until the afternoon of the second day.

During this long period no one is permitted to sleep. There is no music or dancing. Everyone present must perform the tasks allotted him by the shaman. Water has to be drawn from the cenote, wood gathered for the many fires, pits dug and stones collected for a special method of baking, food and drink prepared, and so on. The shaman sacrifices the turkeys and chickens, and ceremoniously cuts the fowl into small pieces on a flat-topped, rectangular stone. Some of the food is cooked in pots resting on three stones over an open fire. Other foods are baked; this is done by placing the food on banana leaves laid over preheated stones inside a pit, covered with more leaves, then earth, and allowed to bake for several hours. According to one informant, before the food is eaten it is blessed by the shaman, who tosses bits of it into the cenote as an offering to the gods. Much drinking accompanies the entire affair.

This ceremony, called "Cha chac," is directed expressly toward the rain gods to assure that rains will come at the required time to benefit the young, growing corn. The informants stated that it is not just a local custom, but all the neighboring towns and villages practice the ritual. Each community has its own particular place in the forest near their village where the ceremony is held. Only the people of Telchaquillo use the site by Cenote Itzmal Ch'en.

Pottery and artifacts were recovered in varying amounts from all excavations in Mayapan. Discussion of the ceramic material is presented by R. E. Smith (pp. 251-256), and of the artifacts by Proskouriakoff (pp. 256-259). Very little pottery trade material came to light during the past season. A type of Fine Orange was the only imported ware that consistently appeared in most of the excavations, though in very small amounts. More widespread trade was evident from the artifacts of stone, shell, and metal. The first included jade, obsidian, and greenstone; the last, gold and copper.

During Shepard's stay in Yucatan two short journeys were made with her to study present-day methods of making pottery. The first trip was to the village of Mama, 18 km south of Mayapan; the second to Maxcanu, a town southwest of Merida on the railroad to Campeche. At both places samples of the raw materials were obtained and the techniques employed by the potters were observed, with the thought in mind that the clays, tempers, and methods still in use might be similar to those found in the preconquest pottery of Mayapan. The Maxcanu potters stated that their tempering material

was quarried locally, but clay had to be brought in by trade from a source near Becal and Tepakan. The potters from the latter towns obtained tempering material from Maxcanu.

Another of the Department's research objectives in Yucatan is the study of archaeological sites occupied at the same time as Mayapan. Historical sources indicate that most of the contemporary towns in northern Yucatan were subject to the political authority of Mayapan. closely the remains of such towns conform to those of the capital is not known. Certain features such as house plans and grouping, types and architectural style of public buildings, styles of sculpture, pottery, and other artifacts may often be observed without excavation. These data serve to indicate with some degree of accuracy the conformity or nonconformity with the type site.

Reports of ruins obtained verbally from the local inhabitants were investigated whenever possible. Those in the environs of Mayapan received more attention during the past season than sites elsewhere in the peninsula. The ruins of Telchaquillo, over which the modern town is built, are 2 km north of Mayapan. Some of the house mounds scattered around Telchaquillo have plans similar to those of Mayapan, but other house mounds and the larger ceremonial structures appear to predate those of the type site. Just 2 km due south of Mayapan a minor group of ruins surrounds Cenote Mateyac, and these belong to the Mayapan Period. At Cenote Itzimkan, 6 km to the northwest, there is another site, the architecture and pottery of which indicate occupation, perhaps continuously, from the Classic through the Mayapan Period.

Mr. Lawrence Roys and Shook returned to the ruins of Ake (see Year Book No. 50,

pp. 237-238) and spent three days in further study of this important site. The combined results of their investigations will be published in a forthcoming report. An outstanding discovery pertaining to this site and to Izamal was made by Sr. Victor Segovia, of Merida. Acting on our suggestion, he undertook the difficult task of following the ancient roadway, or sache, from Ake to Izamal, a distance of approximately 32 km. He thus confirmed the previous surmise that the great road connected these sites. Along the way Segovia noted two previously unreported ruins, Xemna and Sitpach. Xemna, a site of twenty or more mounds, 7 km east of Ake, was connected with the ancient highway by a branch sacbe 9 m wide, even though the ruins lay only 150 m to the north. Farther east, on Hacienda Muycuyche, was the minor site of Sitpach, consisting of a few scattered mounds along the south edge of the sacbe.

POTTERY OF MAYAPAN

ROBERT E. SMITH

During the first month of the 1952 field season R. E. Smith completed the selection of Yucatan sherd collections to be sent to various institutions in Mexico, Guatemala, and the United States (see Year Book No. 50, p. 225). These collections include wares and types classified on the basis of descriptions by Dr. George W. Brainerd and R. H. Thompson. They contain material from Acanceh, Chichen Itza, Coba, Dzibilchaltun, Holactun, Kabah, Labna, Mani, Mayapan, Oxkintok, Sayil, Uxmal, and Yaxuna.

The greater part of the season was spent in investigating Cenote X-Coton, in the southeast corner of Square T at Mayapan (Jones, 1951, C. I. W. Notes, no. 106, map). This location was selected because of the apparent importance of the cenote in pre-

historic times. The position of two ceremonial structures and a major gateway in the city wall in relation to the cenote, as well as the presence of artificial constructions on the floor of the cavern, suggested that the erstwhile inhabitants of Mayapan laid considerable stress on this natural feature.

X-Coton is a jug-shaped cenote with a banana grove on the stretch of floor that lies below the rather large, circular opening. The opening is eccentrically located over the southern part of the cavern, the northern part being roofed by natural rock. The cenote is, or was in ancient times, a source of water.

Of particular interest and prominence was a platform, more or less centrally located on the floor of the cenote, approximately under the northern edge of the opening. This wedge-shaped structure was mounted by a stairway at the narrow western end, the blunt eastern end being rounded. To the north of the platform there descended to a lower level an artificially constructed stairway, almost completely covered by dirt and bat dung. This refuse was cleared away, revealing 18 steps and a clear pool of water at the bottom.

The platform, presumably ceremonial in function, was cleaned and tested but not completely excavated. A trench dug to bedrock at the eastern end of the structure revealed the fact that the foundations were of large and medium-sized stones that included a small amount of pottery of both Puuc and Mayapan Periods (table 1, D-4, -5, -6, -18). These foundations rested on a leveled fill of dirt nearly 1 m deep that also contained Mayapan and Puuc Period sherds, and a sizable representation of Classic Period and Regional Polychrome pottery (table 1, D-8, -11, -15, -16, -17).

Neither from the trench east of the platform nor from a pit sunk in the floor of the cenote at the center of the banana grove (table 1, D-10) did chronological stratigraphy materialize. At all depths there occurred Mayapan Period sherds mixed with those of the Puuc. Surface sherds were collected throughout the cenote to a depth of 10 cm (table 1, D-2). These, as might be expected, were largely of the Mayapan Period.

During the gathering of the surface sherds, two natural recesses, or niches, in the wall of the cenote were encountered and excavated. This resulted in the discovery of a sculptured stone dog and run-of-the-mill pottery in one niche (table 1, D-20) and a sculptured squatting human figure associated with a large quantity of censer fragments in the other (table 1, D-9).

Four caves were noted during the examination of Cenote X-Coton. These occur as subsidiary caverns in the peripheral wall of the cenote. Only two could be excavated, the others being either too narrow or too shallow for anyone but a small child to enter.

Cave I was the more interesting. It had been used for what appeared to be secondary burials, possibly in pre-Mayapan Period times. The cave was deep, extending down to water. The lower section was filled with small and medium-sized rocks. Above this, skeletons were found along with many broken vessels of both coarse and fine wares, most of which were of the Puuc Period style, but an appreciable percentage of which were Classic Period and Regional Polychrome (table 1, D-7). A single whole vessel of medium paste slate was recovered. Since it is presumed that the skeletons together with their pots were added at different times, rocks being tossed in to cover them, it is not surprising that the bones were spread over the cave and the pottery was smashed. The action of dripping water of high lime content tended further to obscure the picture, since it cemented the bones to one another and to the rocks.

A small percentage of Mayapan Period sherds was found mixed with the rest of the pottery and bones. No very satisfactory explanation for the presence of these late sherds has been advanced. It is, of course, possible that one or two Mayapan Period individuals were buried there. Actually, the arm bones of one skeleton, just below the top stones, appeared to have been articulated, suggesting that the last burial was primary.

Cave 2 was of less interest. It harbored a few sherds (table 1, D-14), mostly of the Mayapan Period, and a single skeleton in poor and fragmentary condition. This cave was like Cave 1 in containing a layer of small and medium-sized stones below which the sherds and skeleton were found. It differed, however, in having a deep carpet of bat dung. No bats were noted in Cave 1, but they occurred in great numbers in the second cave.

An analysis was made of the pottery collected from house platforms by Ruppert and A. L. Smith (table 1). Twenty-six platform cuts resulted in only 3628 sherds, an average of about 140 sherds per cut. Two major ceramic periods were indicated, the later and the more abundant (92.6 per cent) being the Mayapan Period, the earlier (4.5 per cent) the Puuc Period. Elements of two other ceramic periods were present: the Classic Maya, represented by Regional Polychrome (0.03 per cent), and the Toltec, exemplified by Fine Orange and Plumbate wares (0.53 per cent). Only 2 Plumbate sherds were found at Mayapan, whereas there were 83 specimens of Fine Orange, mostly of a new Mayapan type but with rare occurrences of X and Z.

The excavation that produced the purest Mayapan Period remains was made in Structure J-50-a, where, save for one Fine Orange body fragment, all sherds belonged to the Mayapan Period (table 1, A-13). This cut penetrated a midden which contained, besides pottery, all sorts of bone, shell, obsidian, and stone implements and ornaments (see p. 244). Only one excavation in a house platform yielded a greater percentage of Puuc than of Mayapan Period pottery (table 1, A-12). This was in Structure J-49-a. The entire platform had an unusually large percentage (40.4) of Puuc Period sherds. It would seem from the limited information so far available that house platforms at Mayapan normally were late constructions containing Mayapan Period sherds in the fill. When Puuc Period pottery is the more abundant, as in the above instance, a nearby refuse dump or occupation area of that period is indicated.

On the suggestion of Ruppert and A. L. Smith, a trench was cut into the north side of Structure J-49-b, a house platform. The trench advanced at bedrock level, encountering first the north terrace wall, then a rough retaining wall, and finally ending against a plastered tomb without skeleton or furniture. There being no obvious strata, three more or less arbitrary levels were maintained throughout the trench. The ceramic findings in these levels and between walls did not vary appreciably (table 1, A-6, -7, -11, -18, -19, -20), the percentages of sherds from all the cuts being as follows: Mayapan Period 95.7, Puuc Period 2.1, unidentified 1.4, Fine Orange o.8. Thus, our attempt to find significant stratigraphy was not successful.

A tabulation by periods or wares of the pottery from four city gates and from several ceremonial structures excavated by Shook is included in table 1. Two cuts

TABLE 1
SHERD COUNTS AT MAYAPAN, 1952 SEASON

Cut	Mayapan Period	Puuc Period	Classic Period and Regional Polychrome	Unidenti- fied	Fine Orange	Plumbate	Formative	Total
			Нот	JSE PLATFORM	ıs		<u>'</u>	
A-1	76							76
A-2	189	7		3	2			201
A-3	75	 			 			75
A-4	35							35
A-5	233			18	1	1		253
A-6	42	1		3	1			47
A-7	162			2				164
A-8	45	10						55
A-9	87			11	2			100
A-10	34	2						36
A-11	165	2						167
A-12	34	82		5			1	122
A-13	775				1			776
A-14	31							31
A-15	174				1			175
A-16	205			7	1			213
A-17	118			2				120
A-18	398	16		9	8			431
A-19	155	1						156
A-20	182	4		2				188
A-21	3							3
A-22	65	32		2				99
A-23	26							26
A-24	14	8		4				26
A-25	34		1	10				35
A-26				18				18
Total	3,357	165	1	86	17	1	1	3,628
Per cent	92.6	4.5	0.03	2.3	0.5	0.03	0.04	
				CITY GATES				
B-1	238	3		64				305
B-2	88	7		36	9			140
B-3	134				,			134
B-4	41	36		9				86
B-5	54	34	1	28				117
Total	555	80	1	137	9			782
								702
Per cent	71.0	10.2	0.1	17.5	1.2			

(Continued on following page)

TABLE 1—Continued

Cost	Mayapan	Puuc	Classic Period and	Unidenti-	Fine	Plumbate	Formative	Total
Cut	Period	Period	Regional Polychrome	fied	Orange	Fiumbate	Formative	Total
CEREMONIAL STRUCTURES								
C-1	179	5		28	4			216
C-2	472	16		66			· · · · · · ·	554
C-3	1,185	88	1	85	5	1		1,365
C-5	56							56
Total	1,892	109	1	179	9	1		2,191
Per cent	86.3	4.9	0.1	8.2	0.4	0.1		
			CEN	оте Х-Сото	4			
D-1*	3				3			6
D-1 D-2	2,014	214	8	18	3		1	6 2,255
D-3	516	63	6	9	2		1	596
D-3 D-4	267	142	8	1	2			420
D-4 D-5	588	78	4	1	2			670
D-6	614	213	10	29	3			869
D-7	49	1,049	165	30	3			1,293
D-8	29	18	60	57			1	165
D-9	785	127	3	31			1	915
D-10	52	30	2	14				98
D-11	25	30		11				36
D-12	2,080	37	9	716	34			2,876
D-13	790	235	21	343	3			1,392
D-14	125	9		13				147
D-15	5	13	4	6				28
D-16	11	3	4	6				24
D-17	22	5		47				74
D-18	54	23	5	6		1		88
D-19	95	1		143	1			240
D-20	273	32		33				338
Total	8,397	2,292	309	1,482	48		2	12,530
Per cent	67.0	18.3	2.4	11.8	0.4		0.1	
					1			

^{*} In natural sink in Square K.

associated with Gate T produced unusually high percentages of Puuc Period sherds (table 1, B-4, -5), whereas the ceramic material (over 50 per cent censers) from the near-by ceremonial structures is preponderantly Mayapan Period (table 1, C-2, -3, -5).

The work of the past season at Mayapan leaves us with an intriguing ceramic picture and a serious problem. So far there is every indication that two major ceramic periods existed at the site: the Puuc and the Mayapan. Just where the Puuc-style pottery fits into the chronological sequence is not clear, owing primarily to the lack of recognizable stratigraphy and to the fact that Dr. G. W. Brainerd's extensive study of peninsular ceramics is not yet available. Together with the Puuc Period pottery are frequently found types called by Brainerd Regional Polychrome. This pottery is the same macroscopically as that found in the Peten in both the Early and Late Classic Periods, but more often in the Early Classic. Fine Orange sherds, some 83 in all, plus 2 Plumbate fragments, suggest a Toltec horizon, although the chronological implications of the former ware are still far from clear. If Mayapan had been occupied in Toltec times, however, there should be more wares than Fine Orange and Plumbate to represent that period. Is it possible that a portion of the seemingly Puuc-style pottery is actually of Toltec Period, or should certain Mayapan Period ceramics be assigned to Toltec times? The latter seems the more likely, but much investigation is necessary before the situation will be clear.

Sculpture and Artifacts of Mayapan

TATIANA PROSKOURIAKOFF

Although according to native histories Mayapan was "founded" by the hero Kukulcan, whose name is associated with the

cult of the feathered serpent, even from surface remains it is clear that in fact the city is older, and that long before its reputed "founding" it was occupied by a people with traditions like those of the Puuc. No building of this earlier period has been identified, but everywhere, in surface debris and built into walls of cruder masonry, one can find accurately cut and smoothly faced stones, as well as carved elements of decoration almost identical with those of the most elaborate buildings at Kabah and Uxmal. These stones are cut from carefully selected, smooth, hard limestone, and the relief of their carving is still sharp and clear. There are elements of masks and carved moldings, spools, crosses from the lattice motif, and other forms familiar in the Puuc. In addition, there have been found three excellently carved human heads, distinctly Maya in their features, and designed to be tenoned into masonry construction.

Fragments of stelae with inscriptions, of which there are several, also probably belong to this earlier period. Dates have been suggested for some of them, but none are sufficiently legible to merit acceptance. On other monuments, apparently similar in design to those with inscriptions, glyphs are blocked out but left blank of detail. Stela 1, now in the hacienda at Xkanchakan, is the only low-relief stela on which figures can still be made out. In design it differs from Classic Maya monuments. The relief is flat, the detail is executed in lines only, and the composition is divided into panels, one containing glyph blocks, and the other an arrangement of figures suggestive of a scene from a codex. The largest stela of this type lies at the base of a round building on the east edge of the main ceremonial group. It is carved of very poor limestone filled with fossil inclusions of shell; its position, the choice of poor material, and

the omission of glyph details raise the question whether the stela cult did not still survive in the later period of Mayapan.

Most of the monuments associated with the late remains are carved of the same coarse-grained, shell-filled limestone, which erodes unevenly, so that much detail is lost and the original quality of the carvings is difficult to judge. As a group, these sculptures are different from any other known group in Yucatan. So far, no attempt has been made to define the style or to trace its antecedents. The work this season consisted mainly in completing and supplementing the record previously made by Andrews (Year Book No. 41, pp. 261-263) and others of the sculptures, which are concentrated in the main ceremonial group. In the course of this work, the remains of a very low round platform with rectangular additions were discovered in the plaza north of the Castillo. A number of carved monuments and fragments of plain stelae were scattered on and around this platform.

Certain features of late sculpture at Mayapan derive from the Toltec tradition. Among these are carved serpent columns and banner holders in the form of squatting figures. The serpent columns are more crudely executed than those at Chichen Itza. The shaft is plain, and the heads of the serpents are separate elements. The design was probably finished in stucco. The banner holders are animal figures, with heads turned sharply to one side in the manner of Chacmool statues. No Chacmool has been found, but perhaps this is not significant, for at Chichen Itza they were often buried in construction. Mayapan, however, clearly lacks the rich low-relief carvings of warriors, priests, and processions, and the carved altars and atlanteans that distinguish Chichen Itza. Two low-relief figures of monkeys, found

on a mound outside of the main group, are of crude workmanship. A very late date is suggested for them by what looks like the Mexican sign Tecpatl, a flint knife with the features of a skull, behind one of the monkeys.

The principal carvings of the Main Group are figures of somewhat less than life size, in full round or in three-quarters relief. Though some are carved on all four sides, they tend to retain a blocklike character, with stronger modeling on the front and sides than on the back. A number have round depressions on top of the headdress. Other figures have a tenon extending part way up the back, as if they were set into masonry. One such figure was found on top of a mound in a position which implied that it once stood in a shrine. These figures are clearly not stelae, nor can they be regarded merely as elements of architectural decoration. They may be the idols which Kukulcan is said to have introduced into Yucatan in late times. Several squatting figures resemble in a general way the numerous idols of Mexico, and one excavated in Cenote X-Coton (see p. 252) is undeniably an idol. The eroded condition of most of the figures precludes a detailed study of their dress and accouterments, which otherwise might furnish a clue to their origin or stylistic affiliations. Both Maya and Mexican traits can be recognized in the surviving detail, but there is not enough to define a complex of traits.

Several other forms seemingly peculiar to Mayapan were observed, but because of the very poor condition of the sculptures found on the surface, a full description is postponed in the hope that future excavations may bring to light better-preserved examples to enable us to classify and evaluate the material more fully.

Any sound appraisal of the sculpture of

Mayapan is further hindered by our very imperfect knowledge of the history of sculpture elsewhere in Yucatan. Architectural and ceramic studies indicate that there was a strong and uniform tradition centered in the Puuc region, which at one time spread widely over the northern Maya area. This condition does not seem to be reflected in figure sculpture, which shows bewildering variability. Without a basic chronology of styles to which one can refer, untangling the complex maze of influences and relationships which the Yucatan material implies is a formidable, perhaps an impossible, task. At present, studies limited to variations within definable types or styles of sculpture offer more chance of success than does an attempt to cover the whole field. A classification of mask elements and other architectural motifs, with particular reference to the technique of carving and assemblage, may help us to relate the earlier occupation of Mayapan specifically to individual sites and possibly to a limited period in the Puuc style. A start at gathering together such material was made last fall. A similar study of serpent heads may also prove worth while.

A broader field of interest can be served by a comprehensive study of the low-relief sculptures at Chichen Itza. Although this is the best-known body of sculpture in Yucatan, many of the reliefs remain inadequately recorded, and the material has never been studied as a whole. The absence of comparable sculpture at Mayapan poses interesting questions in regard to the chronological and cultural relations between the two cities. Once the chronology is established through ceramic and architectural sequences, the sculpture of Chichen Itza may become a more valuable source of data on the ethnography and history of Mayapan than are its own inferior sculptures.

The first desideratum is a more complete record than we now have of all exposed low reliefs at Chichen Itza, particularly those depicting human activities. With a view to determining whether a project to obtain such a record is feasible, and what work and equipment it would involve, Pollock and Proskouriakoff spent several days at the site listing sculptures of which it is desirable to have photographs. The estimate of the number of negatives necessary to cover unrecorded or poorly recorded sculpture is approximate, since this number depends on the type of equipment used and the degree of success in obtaining good lighting for extended areas. About 1050 negatives was estimated as the minimum needed to cover the material actually examined. To this should be added perhaps another hundred for sculptures in several outlying structures which were not visited, and a margin for duplication and spoilage. Many of the subjects require night photography with artificial lighting. In a small number of cases, the use of scaffolding seems necessary.

In addition to the bas-reliefs, there are many figures of atlantean type at Chichen Itza not thoroughly recorded. These, being more formal in design, offer fewer ethnographic data. Of more immediate relevance to our studies are minor fullround figures, which have largely escaped attention. It is possible that among aberrant and eroded figures at Chichen Itza and in the Puuc there are some comparable to the Mayapan idols. A systematic search for them would hardly be advisable at this time, but a review of the known material in the light of what is found at Mayapan may give evidence of late occupation where we do not suspect it.

More than 200 artifacts from Mayapan were catalogued this season. Some came from a surface collection made by Morris Jones in previous years, many more were gathered by Bullard this year, and others were found in excavations. Among the artifacts are a number of tools of native limestone, most of them articles of household use, others, such as stones for smoothing plaster, being the implements of artisans in building and other trades. There were 83 chipped flints of various forms, differing widely in quality of workmanship. Most were coated with a deep white patina. Among the specialized forms were two sacrificial knives like those recovered from the Sacred Cenote at Chichen Itza, a number of small hatchet-form and triangular tools of unknown function, and a variety of knives and projectile points. Two very small points may have served as arrowheads, but the only arrowpoint of specialized form so far recovered was a small, delicately chipped point of obsidian, excavated by Shook.

Obsidian occurred in quantity, mostly in the form of thin flake-blades. Its color is translucent gray, often heavily streaked with a darker tone. A used core intimates that the material was imported unworked, and that the flaking was done locally. Most common among other artifacts of imported stone were small polished celts of dark greenstone. Several jade beads and an exquisitely carved small pendant in the form of a human head were discovered by Shook in his excavations.

Other materials included pottery, shell, bone, antler, and coral. Perforated teeth, both animal and human, were apparently used in necklaces. Several very small flakes of gold and a tiny ball of copper, probably from a bell, established the presence of metals (see p. 248). The collection is still too small to represent the whole range of artifacts we may expect to find at Mayapan. The present material, therefore, after being catalogued and, in part, photographed, was stored for future study.

CHACCHOB, YUCATAN

H. E. D. POLLOCK AND GUSTAV STRÖMSVIK

The ruins of Chacchob are in the District of Tekax, some 13 km by road and 10 or 11 km airline southeast of the town of Teabo. The site was first brought to the attention of the public over 100 years ago in an anonymous article that appeared in a Merida periodical of the time. The part of the account that particularly has caught the attention of archaeologists is the statement that the ruins are surrounded by the remains of an ancient wall. Walled sites in the lowland Maya area are rare, and their occurrence is important because of certain implications as to the social structure and the general character of the civilization of the builders.

Although the information provided by the anonymous antiquarian of a century ago has from time to time been made use of by modern authors, the truth of his assertions, so far as is known to the present writers, had never been confirmed by competent archaeological opinion. The work of Carnegie Institution at Mayapan, the largest and most important walled city known in Yucatan, made it of peculiar importance not only to verify the existence of a possibly similar, near-by site, but to know something of the character of the remains.

In May 1951, Eugenio May, a Yucatecan employee who has worked for many years with the Department, was commissioned to visit Chacchob and to report his observations, particularly with reference to any indications of a wall surrounding the ruins. May spent two days at the site, and his report left little doubt but that the ancient settlement had in fact been walled. It was consequently planned that further reconnaissance should take place during the 1952 field season.

Pollock and Strömsvik, traveling by

truck, left the Institution's base camp at Telchaquillo February 27. The road, the old highway to Tekit, ran southeast through the ruins of Mayapan. Just short of Km 12, and approximately 10 km from the center of Mayapan, the site of Chumul was noted at the left of the road. This moderate-sized group of ruins, which was visited on our return journey, rests on a hill. The principal pyramid is clearly seen from Mayapan, and because of its natural elevation gives the impression at that distance of being a much larger structure than it is. This pyramid is flanked by lesser pyramidal structures, one on each side, and several mounds of indeterminate character complete the group. Although all buildings are fallen, the character of the masonry and the stone cutting, the architectural decoration, and certain monoliths mark the ruins as of the Puuc Period. Features observed include finely cut lintels and fullwidth jamb stones, colonnette façade elements, tapered columnar (picote) altars, and a rectangular altar.

Passing through Tekit (Km 21), a sizable town, our road continued south-southeast toward Chumayel and Teabo. At Km 25 we reached Hacienda Ukum, where there are ruins and where May in the previous year had discovered a sculptured stone built into the floor of the abandoned machine house of the hacienda. This stone, which was photographed on our return trip, appears to have been a door jamb. It carries a rather typical Maya figure, erect and with plumed headdress, facing a vertical row of glyph blocks and standing above two horizontal rows of glyph blocks. The 13 or 14 hieroglyphs represented have not been deciphered. The sculpture and the character of the jamb stone suggest this to be a work of the Puuc Period, an impression strengthened by the presence of certain architectural elements (spools)

typical of that period. A superficial view of the near-by ruins disclosed them to be no more than mounds of stone, but betterpreserved structures may lie hidden in the forest.

Chumayel was passed at Km 34, and Teabo, a town of about the same size as Tekit, at Km 38. Teabo clearly was once an aboriginal settlement. A huge mound of rock, a large city block in area, from which much of the present town probably was built, gives ample testimony to the size of the structure that has withstood four centuries of quarrying and still remains a massive bulk of stone. Across the street from this great pile are the ruins of a large and once beautiful convent of the colonial period.

Leaving Teabo, we continued southeast on the road that eventually reaches the village of Xaya. At Km 48 we reached Hacienda San Diego Max, once a henequen plantation, now turned over to cattle raising. The hacienda is the property of Jose Dolores Salas of Teabo, who most kindly offered us the use of his buildings, apologizing for their roofless condition. Here we made camp, our information being that this was as far as the truck could go. It was later discovered that with a small amount of clearing the road was passable as far as Chacchob. Our entire journey from Telchaquillo to San Diego Max consumed approximately 4 hours and 30 minutes (running time), of which I hour and 40 minutes were devoted to the 10 km from Teabo to San Diego Max. It was subsequently determined that the additional 3 km to the ruins was a trip of 20 minutes by truck.

Chacchob, now the site of a small cattle ranch, lies east-southeast of San Diego Max, on the road to Xaya. The ranch and the ruins are reported to be on the lands of the latter village. Previously this terrain

was a part of Hacienda San Diego Max, and the corral walls, which include many stones from the ruins, and the masonry curb of the well are unquestionably products of the period of hacienda ownership. This well is worthy of note. At first sight it appears to be of artificial construction, typical of the innumerable wells dug during the colonial period and in later times. On closer examination, however, it is apparent that only the curb and a small depth of fill below are man-made. The well proper is a natural bottle-shaped opening reaching down to water, in other words, a cenote. Deep rope cuts in the rim of natural rock below the artificial fill indicate centuries of use and almost surely date from the period of the ruins. This was the only source of water observed by us, or reported by the natives, within the area of the ruins. We did, however, hear of other natural water sources not far distant.

The ruins of Chacchob, which are surrounded by a wall, occupy a relatively small area covered for the most part by low, scrubby brush. Our explorations were confined rather closely to the territory within the wall, but there is no reason to believe that structures of any size lie beyond that limit, although house mounds may extend an indeterminate distance in any direction. The area outlined by the wall is a rough oval, some 500 m on the long axis, which runs east-southeast and west-northwest, and approximately 350 m shorter dimension. through the wall were noted at the northeast, southeast, and southwest, with reference to the center of the oval, and what probably had been a passage was observed at the northwest.

The ranch and well lie almost at the center of the ruins. Immediately to the north of center are two pyramidal struc-

tures, and a similar structure is just to the south. These range in height from possibly 6 m to 10 m, the highest being the northernmost. Farther south is a large, low rectangular platform, some 50 by 60 m, with a fallen, once vaulted building on the western edge. It is this building and the pyramid south of the ranch that have furnished much of the stone for the corrals and other postconquest construction. In the eastern part of the site, approximately on the long axis, is the largest structure in the ruins, the so-called Castillo. An irregularly shaped platform that supported the remnants of once vaulted structures as well as house platforms occurs in the western part of the area. Remains of house platforms are scattered throughout the site, their number and concentration not being known.

Five days were devoted to exploration and study of the ruins. During that period data for a sketch map were gathered, architectural details were recorded, and a small amount of pottery was collected. No sculpture of consequence was noted.

The wall surrounding the site was a matter of primary interest. It differs from the wall of Mayapan in that it follows a course clearly selected to utilize, wherever possible, natural elevations of the terrain. This is strikingly apparent in the southeastern, southern, and western sections, where the wall follows natural ridges several meters in height for a distance that comprises something like two-thirds of the circumference. On the other hand, the Chacchob wall is very similar to that of Mayapan in design and construction (see p. 247). It consists of an inner bench and a higher, outer parapet, the whole built of rather large, roughly shaped stones. The parapet is for the most part formed of large stones set on edge, and generally does not rise more than 30 or 40 cm above the

bench. Remains of a plaster floor on top of the bench at one point clearly indicate how that part was finished, and it is entirely possible that all surfaces once were plastered.

The total thickness of the wall would seem to average about 4 m. It is difficult to give any estimate of the present maximum height because of the confusion, without excavation, between what is natural and what is artificial. It would seem reasonable, however, to guess that the parapet attains at places a height of at least 2 m above ground. Maximum total height, including natural elevation, must be at least 6 m, probably more. Gates are simple cuts through the wall, the best preserved, those at the southeast and southwest, being about 2 m wide. Steps are visible leading up to these gates, this being made necessary by the natural rise.

The only structure sufficiently well preserved to offer any appreciable amount of architectural detail is the Castillo. This large complex of terraces and adjoining courts covers an area approximately 60 by 100 m. The principal and dominating building is a pyramid which rises in two great terraces to a height of 13-16 m above ground level, the difference in elevation being due to the natural slope of the terrain. No trace remains of the temple building that once must have crowned the pyramid. The structure faces west, as is shown by the stairway that rises from a courtyard at the foot of the pyramid. This courtyard is formed by ranges of vaulted rooms, some of which are still standing, that extend west from the northwest and southwest corners of the lower terrace of the pyramid. To the south of the pyramid and of the courtyard are other courts and terraces, some of which supported vaulted buildings now fallen.

There is good reason to believe that this large architectural complex grew by accre-

tion. Construction may thus have extended over considerable time, and more than one cultural period may be represented. The fact nevertheless remains that all architectural details well enough preserved to form reliable criteria point to the Puuc Period. The vaulted rooms at the western foot of the pyramid are unquestionably of Puuc style, and there are indications that the pyramid was built contemporaneously with these rooms, although this is by no means certain.

Nowhere else at the site was there noted standing masonry that would indicate the period of construction. There were, however, many examples of loose building stones which by reason of design and style of workmanship could be associated with the Puuc Period. Only the wall around the site, because of its similarity to the wall of Mayapan, and a somewhat aberrant group of house platforms suggested any other period of occupation.

A number of structures that appeared to be the remains of dwellings were observed. These occurred both on platforms that supported the debris of vaulted buildings and on platforms with dwelling-type remains only. The characteristic plan is that of a rectangular room, or a series of two or three rectangular rooms, outlined by low stone walls one or two courses high. Doorways to rooms were frequently marked by jambs of larger stones. These low walls presumably were the foundations for houses of perishable materials. There also were instances of simple platforms which showed no surviving traces of houses but which might well have supported perishable construction. These house platforms occurred singly on a larger platform or terrace, or in groups of two or three that formed a small courtyard.

At no place was there noted the typical Mayapan type of dwelling with benches, nor was there noted an instance of a rear group of houses with low walls that employed much cruder stone work than all others seen suggested a different period of construction, but there was nothing to suggest a similarity to Mayapan. On the whole, the dwellings we observed approximate more closely the low walled remains, presumably domiciliary, that are found in the Puuc area.

During our last day at Chacchob time was available to dig one trench in search of pottery. This was located near a house platform. Soil was found to a depth of 40-50 cm before bedrock was encountered. Two arbitrary levels were maintained, each about 20 cm in depth. Only 165 fragments of pottery were recovered. R. E. Smith examined this material after our return to Telchaquillo. Thirty-six of the sherds were too eroded or too fragmentary to be identified; the remainder were typical of the Puuc Period. Nothing was noted that might be considered intrusive in Yucatan, nor was there a single sherd that might be suspected of belonging to the Mayapan Period. There were no significant differences in the pottery from the two levels in the trench.

We returned to Telchaquillo on March 4, having devoted five days to our reconnaissance of the ruins. The results of our work may quickly be summarized.

Chacchob is a small site surrounded by a wall. In many respects the wall is similar to that which encircles the ruins of Mayapan. On the other hand, the architecture of Chacchob, or such of it as was observed, both formal and domestic, seems to be of the Puuc Period. This time of occupation is further attested by the small sample of pottery recovered. As walled sites of the Maya lowlands are commonly considered to be of a period later than that of the Puuc, and roughly contemporaneous with the major occupation of Mayapan, these

findings were unexpected. Interpretation of these observations, however, should proceed with caution. Our evidence is fragmentary, and there certainly remains the possibility that Chacchob underwent a late period of occupation, traces of which were not seen or not recognized by us. The wall around the site may represent that period, but in such case there should be other, contemporaneous remains. Before any conclusions are reached, it would be well to have a better knowledge of the house platforms and a much larger sample of the pottery.

CERAMIC TECHNOLOGY

ANNA O. SHEPARD

Laboratory work on the technology of Yucatan pottery was supplemented by a trip to Yucatan during the latter part of April 1951. A week at the Telchaquillo headquarters of the Department enabled Shepard to make a microscopic examination of sizable samples of Mayapan and Puuc Period pottery recovered this season, to search for clay deposits in the cenotes and sascab pits of the site, to visit the pottery-making village of Mama, and to attend the staff conference held at the end of the field season. The second week in Yucatan was devoted to the study of collections at the Merida office. At this time Shepard also held conferences with R. E. Smith and Shook on the code which will be adopted for the pottery punch-card file for the Merida office, and spent a day at Maxcanu observing potters (see p. 250).

Not the least important advantage of the trip was the opportunity it afforded Shepard to work with R. E. Smith and Shook. The technological study of pottery is necessarily a co-operative project between the specialist and the field archaeologist who collects the material, knows its context, and formulates questions regard-

ing it. The more the field archaeologist knows about technological methods, their requirements, possibilities, and limitations, the more effectively the facilities of the ceramic laboratory will be used. Familiarity with technological methods is best gained by observation of work in progress and actual participation in those phases of the work that do not require special laboratory techniques, one of the most important of which is preliminary paste classification with the binocular microscope. During Shepard's stay, Smith and Shook gained some familiarity with the characteristics of the different paste types and their range of variations, and a set of reference chips of typical and distinctive pastes was prepared for them.

Knowledge of the distribution and properties of clays and nonplastic materials of Yucatan is essential to a full understanding of the ceramic development of the region. The materials used by present-day potters afford a practicable and convenient introduction to this subject, and their study has a direct bearing on questions of continuity of ceramic tradition. In September R. H. Thompson spent a week at the laboratory on his return from Yucatan and left his collection of potters' materials for analysis (see p. 266). Since the geological formations of northern Yucatan are uniform, and the samples were collected from eight villages, from Valladolid on the east to Campeche on the west, these clays promised to give some indication of the types of material available to prehistoric potters. We were interested in the extent of variation in the properties of these clays, how they might affect the potters' techniques, whether or not montmorillonitic clays are common, since there is evidence that slate ware slip is of this type, and whether or not any clays now being used are comparable to that of slate slip.

The clays are residual from limestone. Some are calcareous, others noncalcareous, but all are very fine-textured and highly plastic. Their mineralogical composition was studied by means of differential thermal analysis, X-ray diffraction, petrographic examination, and staining tests. The preliminary results indicate that kaolinitic clays are more common than we anticipated, although at least one montmorillonitic type is represented in the series. Firing tests show that all except two of the clays are red-firing, and none of them retains carbon paint effectively. Since none is comparable to slate slip, the latter is apparently not a common type of clay in Yucatan, and it may have been traded extensively in prehistoric times.

The tempering materials collected by Thompson are all calcites, but they differ in form, in degree of crystallinity, and in purity. A matter of interest is the percentage of temper used by potters and its adjustment to the requirements of the clay. Chemical determination of the calcium carbonate in the clays, in the tempered pastes, and in the insoluble residues of the tempers showed that the percentage of temper is uniformly high, but there is no evidence of adjustment of amount to compensate for either a calcareous clay or an argillaceous temper.

During the year, the detailed petrographic analysis of Yucatan paste was carried forward, together with the preparation of a punch-card index of thin sections to facilitate the study of distribution of types and correlations of paste and style. The four paste classes represented by Yucatan pottery (sherd, calcite, volcanic ash, and lumpy clay) are clearly distinguishable and easily identified; our principal task has been to determine whether or not there are varieties within them that are localized. A question of primary interest with regard

to the ash temper is whether or not more than one source of supply is indicated. All thin sections of this paste have been reviewed in some detail with this question in mind. The ash in all these sections is comparable in form and in sparseness and fineness of mineral inclusions. The paste is distinguished by the occurrence of particles of tuff or indurated ash. The similarity of these examples suggests derivation of the ash from a single source or from similar formations.

As yet we have no clue to the occurrence of this type of ash in Yucatan. If sherd collections were available, the geographic differences in frequency of occurrence might narrow the likely area of its origin. The reference collections selected from pottery provided by previous explorations in Yucatan, and now stored in Merida, were examined with this question in mind, but they proved to be too reduced to give statistical results. As a tempering material volcanic ash is far superior to calcite, which, aside from the grave disadvantage of limiting firing temperature in consequence of calcination and hydration, gives a granular texture unless it is very fine. One of the puzzling questions of Yucatan ceramic history is why volcanic ash temper fell into disuse. Possibly the source of supply was cut off, or pottery-making in the centers using ash was interrupted by political disorders and the custom was lost. Our first problem is to establish the time of its disappearance.

Calcite, everywhere available in Yucatan, is the one tempering material that has been used continuously up to the present time, and it shows a great variety of types. Present-day potters are using two and sometimes three types, either mixed or for different classes of vessels. R. H. Thompson reports that the potters think certain types are superior for cook pots,

an idea to which thermal tests have given no support. There is no reason to suppose that conditions were simpler in prehistoric times, but the homogeneity and distinctiveness of certain calcite-tempered pastes and their association with particular pottery types, or their limitation to certain regions and periods, leave no doubt that there was consistent selection. Only by systematic study of large and well distributed collections can the significant variations be recognized. To date a number have been defined.

The pastes of all sherds recognized as intrusive in Yucatan are distinct from the indigenous pastes. Also, a number of foreign pastes have been found among nondescript sherds. We are handicapped in establishing the source of these intrusives because of inadequate data on the pottery of regions with which Yucatan may have maintained trade. We are therefore attempting gradually to build up knowledge of distribution as collections become available for study. Four such collections were examined during the year: Dr. Wells Jakeman gave us the privilege of examining and preparing thin sections of his material from Aguatal near Laguna de Terminos; Dr. Gordon Ekholm gave us a series of representative types from Panuco; Dr. Eduardo Noguero supplied us with red-onbrown sherds from Xochicalco which Dr. George W. Brainerd suggested should be compared with Yucatan red-on-slate; and, while Shepard was in Merida, Dr. Alberto Ruz gave her the opportunity to examine his Etzna collection, which is of interest in relation to Peten types.

Ceramic studies carried on in the laboratory generally raise questions regarding trade in pottery, its effect on the spread of techniques and styles, and its reflection of contacts between peoples. Often the material required to answer such questions can be obtained only by extensive survey and testing. It is therefore necessary to formulate our problems directly with reference to the materials which are being recovered through field operations. The work at Mayapan has not yet progressed far enough to outline broad technological problems, but acquaintance with field conditions and the relations observed among present-day potters of the region have clarified the problems of regional exchange in Yucatan and given us a sounder working basis.

Modern Maya Pottery of Yucatan Raymond H. Thompson

During the late spring and the summer of 1951 R. H. Thompson made a study of pottery-making among the present-day Yucatec Maya. Previous studies of this subject have emphasized the uniformity of the method of pottery manufacture in this area and have singled out the use of the kabal, a foot-turned, unpivoted wooden support for the vessel, as the most striking feature of the method. The present study was aimed at documenting as much ethnographic detail as possible in order to accumulate a body of data on pottery-making which could serve as the basis for inference to archaeological ceramic materials.

Every village where pottery was known to be made was visited. These were Uayma, Valladolid, Mama, Ticul, Maxcanu in Yucatan; and Becal, Tepakan, Campeche, Lerma in Campeche. Also visited were Merida and Izamal, where pottery is known to have been made around the turn of the century, but no surviving potters could be located at either place. All the steps in the pottery-making process from the collection of materials to the firing of vessels were recorded. Particular effort was made to obtain a complete photographic record. As much of the

total process as possible was observed and recorded at each village visited. Further information was obtained by interrogation. A fairly extensive Maya vocabulary of pottery terms was compiled. Samples of materials, implements used in the manufacture, vessels, and other pottery objects were collected in all the villages. The samples of clay, temper, paste, and paint were turned over to Shepard, who has begun a technological study of them.

In addition, several old collections of Yucatan pottery and pottery-making equipment were examined. The largest and best documented of these is in the Chicago Natural History Museum. It was obtained from potters in Izamal in 1895 by George F. Gaumer, who set down a very complete description of the pottery-making process as practiced there. There are a number of small collections in the Peabody Museum at Harvard University. The earliest-known modern vessels from Yucatan were collected for the Peabody Museum by Edward Palmer in Progreso in 1878 and by Alexander Agassiz in Merida in 1882. E. H. Thompson made a collection of vessels and pottery-making equipment largely from Ticul in 1900. He also prepared a paper on Maya pottery-making, but failed to separate observation of contemporary manufacture from speculation about the methods employed by prehistoric potters. Another small collection is in the University Museum at Philadelphia. It was made by Henry C. Mercer, who published several short papers on his observations of pottery manufacture in Merida.

AGRICULTURE, ECONOMY, AND POPULATION
DENSITIES OF THE MAYA

Joseph A. Hester, Jr.

Under an Institution Fellowship a study is being made of geographical conditions which could have affected the economy of the Maya civilization, particularly in the State of Yucatan. The intent of this project is to investigate the agricultural possibilities in Yucatan, within a framework of aboriginal technology, with the hope of arriving at a somewhat firmer basis for an evaluation of Maya economy, and perhaps a more realistic estimate of aboriginal population densities.

The artifacts which give substance to archaeology include more than the durable remains of architecture and implements. It is almost inevitable that a culture which has occupied an area as large as the Peninsula of Yucatan, and occupied it intensively over a period of many centuries, should have left its mark plainly upon the region. Though increasing social elaboration, as evidenced in the architecture, mythology, and Spanish chronicles, certainly required the presence of the Maya in urban centers, it also required that they go farther afield in the satisfaction of their consequently heightened economic needs, and that they intensify the technology already at hand. In Yucatan, as elsewhere in the Americas, since there existed little significant mechanical means and no motive force other than man himself, the residue of aboriginal economic life in all probability is confined largely to various aspects of agriculture.

It is abundantly apparent from the literature on Maya archaeology that authorities are by no means in accord in their estimates of population densities during, for example, a period of intensive occupation of the peninsula in pre-Columbian times. It has been held by some authors that the voluminous and often intricate architecture demanded large populations with high density. Others have pointed out that the agricultural potential of the 100,000 or so square miles of the lowland Maya area is much too low, on the basis of milpa agriculture, to support the large popula-

tions postulated on the basis of architectural remains.

All estimates of population density which are projected into the past must depend on the common economic denominator, agriculture. With respect to this factor, it has been held variously by modern investigators that the surviving, and presumably ancient, milpa method of growing maize, the chief staple apparently throughout the time of the Maya habitation of the peninsula, is (1) the most advantageous usage of the rather poor soil, and (2) a ruinous method of cultivation which hastens the destruction of the soil.

Modern Maya agriculture requires relatively few tools and simple techniques, but considerable know-how. The milpa system apparently is wasteful of the natural vegetation, the soil, and presumably the fertility of the soil. Furthermore, it is the practice in Yucatan to plant a field for usually no more than two years and then to move to a new field from which has been cleared ten or more years' growth of natural vegetation. The yield of maize declines in the second year of planting in a given milpa, a phenomenon apparently universal in the lowland areas. This has been attributed to such causes as decline in soil fertility; increase in competition from natural vegetation or "weeds," particularly grasses; inability to kill off insect pests when the field is fired during the second season, because of insufficient fuel furnished by the light growth of the previous year; and loss of soil due to erosion of the exposed milpa during the first year.

Several contradictory observations regarding the apparent decline in soil fertility are under investigation. One is that in the northern and western parts of the State of Yucatan, where soil generally is thin, seemingly poor, and very rocky, henequen plantations are maintained year

after year, sometimes in increasing production, and are replanted almost as soon as the old plants cease to be productive. Occasionally, in the three years or so before newly planted henequen shoots come into production, maize is seen growing between the rows of henequen. In other instances henequen plantations are adjacent to productive milpas. It seems to be obvious that henequen and maize do equally well on the same type of soil, and that henequen is not subject to decreased productivity due to prolonged use of the soil. A second contradiction may be that the natural vegetation does not seem to be noticeably affected by diminished fertility. By the second rainy season after a milpa has been abandoned to return to "bush," the soil is supporting a heavier growth of natural vegetation than it ever supported of maize.

It is hoped that the information gained by studying the agriculture and agricultural lands of the present-day Maya, added to the considerable data already published, will provide the basis for a study of Maya economy in aboriginal times. There are several ways in which this study is being approached. The first and most obvious is the ethnobotanical one. This phase is being pursued at Telchaquillo with the assistance of Daniel Vargas, a milpero with an intimate and extensive knowledge of food and medicinal plants in that area.

In pre-Columbian times the New World was rich in quality and quantity of domesticated plants. Mexico was one of the more favored regions in this respect. Modern Mexico is vastly more wealthy in diversity of agricultural products, largely because of the importation and adaptation of Old World plants. A basic assumption is being made in this study that, in general, no Old World plant has replaced a New World plant of the same type (e.g., starch plants) in any given locality unless the

Old World plant offered some decided advantage. That is, the Old World plants for the most part now occur in Mexico as additions to, and not as replacements for, the pre-existing New World plants. Another, and perhaps self-evident, postulate is that the native American plants, particularly those which are important staple crops, were, for the most part, long adapted to the area in which they occurred at the time of Old World contact. Consequently, with proper discretion, certain plants, such as those of dietary staple rank, may be regarded as archaeological indices, less subject, in some ways, to outside influence than are monumental architecture and ceramics. Domesticated plants have a further advantage over inanimate objects in that they may be subjected to genetical analysis.

The ethnobotanical collection that is being made is expected to furnish further insight, once identifications have been completed and the Old World plants subtracted from the New, not only into aboriginal economics, but perhaps into the problems of prehistoric population movements as well, provided in the latter instance that the centers of domestication of the various plants are reasonably well established.

A second approach to the agricultural problem comes through a limited study of the properties of the various soils of the peninsula. The area covered is largely the land bordering the paved and otherwise passable roads to which access may be had from Merida. Soil specimens are being gathered every 10 km along the highways, wherever possible, and at archaeological and modern sites—in any event, at places which can be located readily on an accurate map. The purpose is to discover any soil types beyond the tolerance of maize. Conclusive tests have not been accomplished at this writing, but prelimi-

nary values, obtained in pH tests of the specimens already collected, indicate that few, if any, of the soils are too acid or too alkaline for maize cultivation. A probable exception appears to be the coastal sands within a few meters of the ocean. Ultimately it is expected that the soil samples will be subjected to a more thorough analysis.

A third approach is being made through a study of available water. This aspect goes farther than agriculture alone, for water is even more intimately associated with man's existence than is agriculture. The rains which support milpa agriculture were the limiting factor, ultimately, in the size of any aboriginal population on the peninsula, since they were the absolute limit upon the food supply. But availability of water during the dry season may have been the limiting factor in the ability of the Maya to realize the full production of their agricultural technology; thus, domestic drinking water is an important factor in the ecological relations between the Maya and their habitat.

It was hoped that during the course of this study some relic of intensive agriculture would be found which would indicate that the larger cities of the Maya were not exclusively dependent on milpa agriculture. Some of the highest pre-Columbian cultures in the New World featured some form of intensive agriculture; for example, the elaborate and highly efficient terraced gardens of the Peruvian Andes, and the chinampas, or so-called floating gardens, of the Valley of Mexico, both of which continue in use in those areas today. The Peruvian terraces, particularly, are unmistakably artificial, and clearly bespeak the agricultural, as well as the economic, intensity of the cultures which built them. They are often the product of considerable engineering and architectural skill, and

have left a lasting impression on the landscape, something which the archaeologist can "sink his teeth into."

At the end of four months in the field, intensive agriculture, particularly of the post-rainy-season variety, remains to be found in Yucatan, in the archaeological sense. It exists in the dooryards around present-day Maya huts, supported usually by water from wells, most of which were not in existence in aboriginal times. Terracing occurs on a minor scale. Occasionally a few cubic meters of soil are walled up crudely on the side of a cenote to allow a papaya tree to take root. What appeared to be agricultural terraces were observed at several places, but investigation indicated that these, in each case, were architectural remains which coincided by chance with a present-day milpa, and thus far none has appeared to have had a clearly agricultural origin.

Water supply is an important item in intensive agriculture. The Andean terraces in many cases derive their water ultimately from glaciers and snow fields; the chinampas are lacustrine phenomena. If evidence of intensive agriculture exists in Yucatan, it is to be sought near perennial sources of water. With this in mind, many cenotes and aguadas have been visited. None has proved of sufficient size to have contributed significantly to the economy of more than one or two families at most. The lakes of Coba, the laguna of Chichancanab, and others in Quintana Roo and southern Campeche have not been visited. They are at least potential sources of information with respect to the question of chinampa agriculture or related techniques.

Two weeks were spent at Telchaquillo in March, and a similar period in May, in an examination of present-day agricultural methods and the question of water supply. Milpas were visited both within and outside the wall of Mayapan. Parenthetically,

it should be noted that the soil in and around Mayapan appears thin, rocky, and of little value agriculturally to the local milperos, who frequently rent land for milpa in the town lands of Tecoh and Xcanchakan. A trip was made to eight socalled aguadas some 15 km south of Telchaquillo and to several others about 8 km to the northeast. The "aguadas" appeared to be large, shallow cenotes rather than true aguadas. The essential difference between aguada and cenote seems to be that the water in the aguada does not communicate readily with the local water table, whereas the surface of the water in a cenote is, in fact, the water table. It was decided on the basis of preliminary inspection that two or more of the eight "aguadas" to the south could have been significant in dry-season agriculture of an intensive character, but that their influence on the dietary of even the local peoples could never have been of any considerable importance. A quadrangle of ruins in total disrepair was located adjacent to Xtantani, the largest of the "aguadas" immediately south of Telchaquillo.

Visits were made to Kabah, Uxmal, and Chichen Itza for a preliminary survey of what the agricultural problems may have been in those areas. It seems probable that agricultural conditions in the three regions differ to an appreciable extent in terms of soil characteristics, and that although the same techniques of cultivation may have been applied to all these areas in archaeological times, local conditions may have affected yields or even types of plants cultivated.

An attempt was made at each site to observe what the settlement pattern may have been, and to determine whether or not domestic dwellings were located within, or immediately adjacent to, the ceremonial centers, as presumably was the case at

Mayapan. Within a 1.5-km radius of Kabah remains of small structures, which conceivably could be construed as house mounds, were noted, but at Kabah as well as at Chichen Itza the vegetation was too heavy for adequate observation. About Uxmal there are extensive cleared fields, frequently, perhaps predominantly, with deep soil that, with care, probably could support a very heavy crop of maize over a long period of time. Observation here was much simplified, and it was noted that the lowest-lying lands usually contained deep soil in which no artifact other than an occasional, and perhaps recently deposited, sherd occurs. All architecture seemed to be confined to higher ground. The frequent hillocks usually supported some vestiges of building, but in each instance the architecture was judged to have been more elaborate than that of the majority of the presumed house mounds at Mayapan. The hillocks often contained near their summit some indication of a chultun, in or about which usually was noticed a quantity of weathered ceramic fragments. No remains whatsoever were detected of a "working class" domicile. It may be noted here that the renowned aguada, usually to be found a kilometer or so south and west of the main ruin group, dried up following the rainy season of 1951 and remained dry through at least the month of May 1952. The three preceding years are reported as having been years of subnormal rainfall.

Kempton, Emerson, Steggerda, and Stadelman, among others, have investigated and published on many of the problems of milpa agriculture. In the present study, it is hoped to extend their investigations into the problems of clearing the milpa with the known aboriginal tools, i.e. without the use of metal, and of the amount of work required to keep the field clear, particularly during the height of the

rainy season, as compared with that involved in present-day "machete" cultivation. In connection with this study of aboriginal milpa technique, and as an examination of soil fertility, soil exhaustion, and the absolute amount of soil required to permit a stalk of maize to grow to maturity, a series of tests is being conducted in which the volume of soil is precisely measured beforehand, the plant being exposed otherwise to conditions as close to those in the milpa as possible.

HISTORICAL GEOGRAPHY AND DOCUMENTARY RESEARCH

RALPH L. ROYS

The late winter and spring were passed in Yucatan, in an investigation of the boundaries of the former native states at the time of the Spanish conquest, for the purpose of compiling a historical geography covering that period. A number of villages and rural sites on or near these former political frontiers were visited, and especial attention was given to two of these borders. One was the eastern limit of the so-called Province of Ah Canul, which extended from a little above the latitude of Campeche to Punta Kopte on the north coast. The other was the line separating the Province of Ah Kin Ch'el on the west from those of Chikincheel (sometimes called Chauaca) and Cupul on the east. Information regarding the latter was obtained at Temax, Buctzotz, Dzilam Gonzalez, and Dzilam Puerto.

Associated with these investigations was a search for more sites from which people were moved in very early colonial times and concentrated in towns, where they could more easily be supervised (Year Book No. 48, pp. 239–240). Calkini, for example, was little more than a village at the time of the conquest, but it soon be-

came a large and important town where a convent was founded. From a native Maya chronicle we know the names of the former settlements from which it was recruited, but they no longer appear on the maps. This problem was largely solved after a visit of some days at Calkini and a tour of the surrounding country, thanks to the co-operation of the local professor, the mayor, and the librarian. Similar inquiries were pursued at Tizimin with some success, but less than at Calkini.

Owing to the kindness of Mr. Don Drury, it was possible to establish headquarters at Colonia Yucatan in the northeast corner of the state and visit the surrounding area. One trip was made to El Cuyo, where a modern lighthouse surmounts an ancient stone mound of considerable size on the sand dunes by the sea. Not far to the south is Laguna Xuaca, from which the important conquest town of Chauaca took its name. Extensive ruins exist at Kantunilkin in Quintana Roo to the east, including one pyramid containing a descent beneath a stepped roof to a series of very narrow vaulted apartments in the heart of the substructure. Several days were spent at a large ruined site close to Colonia Yucatan, assisting Lawrence Roys in making a preliminary plan of the remains. Although no name is now known for this site, its location and a garbled name on three of the older maps give reason to believe that it was the large and important town of Sinsimato, visited by Montejo during his first expedition into the Yucatan Peninsula in 1527-1529.

During the past year The Maya katun prophecies of the Books of Chilam Balam, Series I has been completed for publication. Some time has been spent in reading the proofs of Conquest sites and the subsequent destruction of Maya architecture in the interior of northern Yucatan.

SOUTHWESTERN PREHISTORY

EARL H. MORRIS

During June of the current year, Morris brought to completion the report upon Basket Maker II sites near Durango, Colorado. An important last-minute development in regard to these sites was the dating of certain series of wood samples from them. The dendrochronological material recovered, mostly in the form of charcoal, had been studied from time to time ever since it was exhumed in 1938 and 1940. A few "cutting" dates, ranging from A.D. 188 to 324, had been established, but the bulk of the samples remained unplaced in time. Early in 1952, Dr. Edmund Schulman obtained a beam section from northwestern Arizona which greatly strengthened the growth pattern for the first two centuries of the Christian Era and extended the master chart from its previous nether limit of A.D. 11 back to 59 B.C. The record provided by this beam made possible the placing of one series of the early Durango wood, in which the oldest "cutting" date was A.D. 46 (Schulman, 1952, Tree-Ring Bull., vol. 18, no. 4). Thus it is proved that the span of Basket Maker II occupation of the Durango region continued from A.D. 46 to some time after 324. There still remains another series of cross-dating wood samples which do not mesh with the master chart at any point. It is to be expected that when material has been obtained to carry the San Juan chronology back to still earlier times, it will be found that the Durango floating series will fall before 59 B.c., thus showing Basket Maker occupation of southwestern Colorado to have begun well before the birth of Christ.

The only date for Basket Maker II from sites other than those at Durango is from Cave du Pont in Utah—A.D. 217 (Stallings, 1941, *Tree-Ring Bull.*, vol. 8, no. 1). Thus

the Durango wood has made a highly important contribution to Southwestern prehistory by confirming the presence of the Basket Maker people in the region 171 years earlier than had previously been known.

Publications

MARGARET W. HARRISON

The editor of the Department's publications records but one new title for the current year, most of her time having been spent on furthering the progress of reports in hand listed last year. Concerning that progress it may be noted:

Excavations at Nebaj, Guatemala (Publication 594), by A. Ledyard Smith and A. V. Kidder, was issued in January 1952.

Chichen Itza: architectural notes and plans (Publication 595), by Karl Ruppert, has been somewhat slowed by conflicting demands on both printer and author. It is now in the final stages of page proof.

Copan ceramics: a study of southeastern Maya pottery (Publication 597), by John M. Longyear III, has been printed for some months and is now at the bindery.

The conquest and colonization of Honduras, 1502–1550 (Publication 598), by Robert S. Chamberlain, is prepared for the printer and ready for composition.

Volume XI of Contributions to American Anthropology and History (Publication 596) is now complete with five papers. Four have been under way for some time: Geologic observations on the ancient human footprints near Managua, Nicaragua (no. 52), by Howel Williams; Mound E-III-3, Kaminaljuyu, Guatemala (no. 53), by E. M. Shook and A. V. Kidder; Conquest sites and the subsequent destruction of Maya architecture in the interior of northern Yucatan (no. 54), by Ralph L. Roys; and The ball courts at Copan, with

notes on courts at La Union, Quirigua, San Pedro Pinula, and Asuncion Mita (no. 55), by Gustav Strömsvik. To these has been added Pottery from Chipoc, Alta Verapaz, Guatemala (no. 56), by Robert E. Smith.

Two large manuscripts are now being edited. Nearly finished is *Pottery of Uaxactun*, *Guatemala*, by Robert E. Smith, which is expected to go to press early in the fall. Recently received is *Basket Maker II sites near Durango*, *Colorado*, by Earl H. Morris and Robert F. Burgh. Four appendices give further detail: "Pictographs," by Helen Sloan Daniels; "Durango skeletons," by Charles E. Snow and William T. Sanders; "Plant material from sites in the Durango and La Plata areas of Colorado," by Volney H. Jones and Robert L. Fonner; and "Animal and bird bones

from the Durango sites," by Hugo G. Rodeck.

Seven papers have been issued in the series Notes on Middle American Archaeology and Ethnology: Further notes on three-pronged incense burners and rimhead vessels in Guatemala (no. 105), by Stephen F. De Borhegyi; Map of the ruins of Mayapan, Yucatan, Mexico (no. 106), by Morris R. Jones; The ruins of Cotio, Department of Guatemala, Guatemala (no. 107), by Edwin M. Shook; A possible early classic site in northern Yucatan (no. 108), by William R. Coe II; Waxen idols and a sacrificial rite of the Lacandon (no. 109) and The introduction of Puuc style of dating at Yaxchilan (no. 110), by J. E. S. Thompson; and Zutugil dugout canoes (no. 111), by S. K. Lothrop.

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E. A. Lowe, The Institute for Advanced Study, Princeton, New Jersey. *Collection and study of paleographical material*. (For previous reports see Year Books Nos. 9 to 35, 37 to 40, and 47 to 50.)

The autumn months of 1951 were spent partly in France procuring the descriptions and photographs needed for the completion of volume VI, dealing with the libraries of France excluding Paris, and partly in Oxford settling the final make-up of the volume. During the spring and summer of 1952 first proofs were received of text and plates of the new volume. At the same time work proceeded on the bibliography of volume VII, which deals with the oldest manuscripts of Switzerland. An article was written on the uncial Gospel leaves attached to the celebrated Utrecht Psalter, in which it was demonstrated that those leaves were written in the Northumbrian abbey of Jarrow. Another and longer paper was finished on the much disputed question of the home of the script known as "Luxeuil." Both papers are accompanied by facsimiles.

Field work on volume VII will be started this year. Most of the time will be devoted to the manuscripts of the famous monastery of St. Gall. Photographs to illustrate this volume will be obtained. The gratifying progress that is being made is due partly to able assistance, but mostly to extraordinary co-operation on the part of everyone on the staff of the Clarendon Press at Oxford.

For report of the work of Ralph W. Chaney, see Department of Plant Biology; of Louis B. Flexner, Arthur T. Hertig, and Chester H. Heuser, see Department of Embryology; of Alfred H. Joy, see Mount Wilson and Palomar Observatories; of Ralph L. Roys, see Department of Archaeology.



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